HILIGHT set on as ''
? b 155, 5, 42, 72, 159, 442, 444

19jun01 16:08:38 User242957 Session D275.2
$0.00 0.057 DialUnits File410
$0.00 Estimated cost File410
$0.10 TYMNET
$0.10 Estimated cost this search
$0.11 Estimated total session cost 0.228 DialUnits

SYSTEM:OS - DIALOG OneSearch
File 155:MEDLINE(R) 1966-2001/Jun W3
(c) format only 2001 Dialog Corporation
*File 155: This file has been reloaded. Accession numbers have changed.
Please see Help News155 for further details.
File 5:Biosis Previews(R) 1969-2001/Jun W1
(c) 2001 BIOSIS
File 42:Pharmaceuticl News Idx 1974-2001/JUN W2
(c)2001 ProQuest Info&Learning
File 72:EMBASE 1993-2001/Jun W2
(c) 2001 Elsevier Science B.V.
*File 72: For information about Explode feature please
see Help News72.
File 159:Cancerlit 1975-2001/May
(c) format only 2001 Dialog Corporation
File 442:AMA Journals 1982-2001/May B2
(c)2001 Amer Med Assn -FARS/DARS apply
*File 442: UDs have been adjusted to reflect the current months
data. See Help News442 for details. PY,PD sort temporarily do not work.
File 444:New England Journal of Med. 1985-2001/Jun W3
(c) 2001 Mass. Med. Soc.

Set Items Description
--- ----- -----------
? s bc1 (w) 2 and treat? and (antisens? or ribozym?)

Processing
Processing
41805 BCL
6452345 2
37486 BCL(W)2
4231658 TREAT?
46939 ANTISENS?
7736 RIBOZYM?
S1 659 BCL (W) 2 AND TREAT? AND (ANTISENS? OR RIBOZYM?)
? s1 and py<1999

Processing
Processing
Processing
Processing
Processing
6850236 1
26279788 PY<1999
S2 5748886 1 AND PY<1999
? s s1 and py<2000

Processing
659 S1

27745106 PY<2000
S3 429 S1 AND PY<2000

? rd

...examined 50 records (50)
...examined 50 records (100)
...examined 50 records (150)
...examined 50 records (200)
...examined 50 records (250)
...examined 50 records (300)
...examined 50 records (350)
...examined 50 records (400)
...completed examining records
S4 180 RD (unique items)

? t s4/3,ab/all

4/3,AB/1 (Item 1 from file: 155)
DIALOG(R)File 155:MEDLINE(R)
(c) format only 2001 Dialog Corporation. All rts. reserv.

10760133 20265280 PMID: 10807000
Human **Bcl-2 antisense** therapy for lymphomas.
Cotter FE; Waters J; Cunningham D
Division of Cancer Biology, Institute of Child Health, London, UK.
f.cotter@ich.ucl.ac.uk
Biochimica et biophysica acta (NETHERLANDS) Dec 10 **1999**, 1489
(1) p97-106, ISSN 0006-3002 Journal Code: A0W
Languages: ENGLISH
Document type: Journal Article; Review; Review, Tutorial
Record type: Completed

4/3,AB/2 (Item 2 from file: 155)
DIALOG(R)File 155:MEDLINE(R)
(c) format only 2001 Dialog Corporation. All rts. reserv.

10753357 98377769 PMID: 9713967
Apoptotic induction in transformed follicular lymphoma cells by **Bcl-2** down regulation.
Tormo M; Tari AM; McDonnell TJ; Cabanillas F; Garcia-Conde J; Lopez-Berestein G
Department of Bioimmunotherapy, The University of Texas M. D. Anderson Cancer Center, Houston, USA.
Leukemia & lymphoma (SWITZERLAND) Jul **1998**, 30 (3-4) p367-79, ISSN 1042-8194 Journal Code: BNQ
Contract/Grant No.: CA62597, CA, NCI
Languages: ENGLISH
Document type: Journal Article
Record type: Completed
The roles of **Bcl-2** protein and the protein ratio of **Bcl-2** /Bax in regulating cell growth in various lymphoma cell lines were examined. A dose-dependent decrease in **Bcl-2** protein expression was observed in the different lymphomas incubated with lipid-incorporated **bcl-2 antisense** oligonucleotides (L-**bcl-2**). Growth inhibition was observed in a transformed follicular lymphoma (FL) cell line, which has the t(14;18) translocation and **Bcl-2** protein overexpression. One of the mechanisms by which L-**bcl-2** growth inhibition is mediated in these transformed FL cells might be through apoptotic induction, because the **treated** cells had an increased apoptotic index and showed the typical DNA fragmentation. These studies indicate that **Bcl-2** protein is critical in the growth regulation of transformed FL cells. L-**bcl-2** did not induce growth inhibition in lymphoma cells not expressing **Bcl-2** or Bax protein. Thus, the protein ratio of **Bcl-2**/Bax may also be

important in regulating the growth of these lymphomas.

4/3,AB/3 (Item 3 from file: 155)
DIALOG(R)File 155:MEDLINE(R)
(c) format only 2001 Dialog Corporation. All rts. reserv.

10587036 20265279 PMID: 10806999
Oligonucleotide therapeutics for hematologic disorders.
Agarwal N; Gewirtz AM
Department of Internal Medicine, University of Pennsylvania School of Medicine, Philadelphia, USA.
Biochimica et biophysica acta (NETHERLANDS) Dec 10 **1999**, 1489 (1) p85-96, ISSN 0006-3002 Journal Code: A0W
Contract/Grant No.: PO1CA72765, CA, NCI; RO1CA66731, CA, NCI
Languages: ENGLISH
Document type: Journal Article; Review; Review, Tutorial
Record type: Completed
During the last decade, the catalogue of known genes responsible for cell growth, development, and neoplastic transformation has expanded dramatically. Attempts to translate this information into new therapeutic strategies for both hematologic and non-hematologic diseases have accelerated at a rapid pace as well. Inserting genes into cells which either replace, or counter the effects of disease causing genes has been one of the primary ways in which scientists have tried to exploit this new knowledge. Strategies to directly downregulate gene expression have developed in parallel with this approach. The latter include triple helix forming oligonucleotides (ODN) and **'antisense'** ODN. The latter have already entered clinical trials for a variety of disorders. In this monograph, we review the use of these materials in the **treatment** of hematologic diseases, particularly myelogenous leukemias. Problems and possible solutions associated with the use of ODN will be discussed as well.

4/3,AB/4 (Item 4 from file: 155)
DIALOG(R)File 155:MEDLINE(R)
(c) format only 2001 Dialog Corporation. All rts. reserv.

10487161 20093826 PMID: 10628339
The involvement of protein kinase C isoenzymes alpha, epsilon and zeta in the sensitivity to antitumor **treatment** and apoptosis induction.
Spitaler M; Wiesenhofer B; Biedermann V; Seppi T; Zimmermann J; Grunicke H; Hofmann J
Institute of Medical Chemistry and Biochemistry, University of Innsbruck, Austria.
Anticancer research (GREECE) Sep-Oct **1999**, 19 (5B) p3969-76, ISSN 0250-7005 Journal Code: 59L
Languages: ENGLISH
Document type: Journal Article
Record type: Completed
In order to obtain additional information on the involvement of protein kinase C (PKC) isoenzymes in the resistance of cells to anticancer drugs and in the induction of apoptosis, we employed **antisense** oligonucleotides to PKC alpha and PKC zeta, CGP 53506, a new inhibitor of PKC alpha, and cells overexpressing PKC alpha, PKC epsilon and PKC zeta. We found that in HeLa cells which express PKC alpha and zeta, down-modulation of either PKC alpha or PKC zeta with **antisense** oligonucleotides induced apoptosis. The PKC alpha selective inhibitor CGP 53506 reduced the proliferation rate of PKC alpha overexpressing NIH3T3 cells more than that of wild-type cells and induced apoptosis, indicating that such a PKC alpha inhibitor may be useful in the **treatment** of tumors overexpressing PKC alpha such as glioblastomas. NIH3T3 cells overexpressing PKC alpha were more resistant, whereas NIH3T3 cells overexpressing PKC epsilon or PKC zeta were more sensitive to **treatment** with cis-platin, adriamycin or

gamma-irradiation compared to parental NIH3T3 wild-type cells. The observed resistance and sensitization corresponded to the extent of apoptosis induced by these **treatments**. Alterations in the expression of p53, **bcl-2** and bax in the PKC isoenzyme overexpressing cells indicate that these proteins may be involved in the different sensitivities of these cells.

4/3,AB/5 (Item 5 from file: 155)
DIALOG(R)File 155:MEDLINE(R)
(c) format only 2001 Dialog Corporation. All rts. reserv.

10480342 20036328 PMID: 10567231
Heat-shock protein 70 **antisense** oligomers enhance proteasome inhibitor-induced apoptosis.
Robertson JD; Datta K; Biswal SS; Kehrer JP
Division of Pharmacology, College of Pharmacy, The University of Texas at Austin, Austin, TX 78712-1074, USA.
Biochemical journal (ENGLAND) Dec 1 **1999**, 344 Pt 2 p477-85,
ISSN 0264-6021 Journal Code: 9YO
Contract/Grant No.: ES07784, ES, NIEHS; HL51005, HL, NHLBI
Languages: ENGLISH
Document type: Journal Article
Record type: Completed
Recent evidence supports a role for heat-shock protein 70 (hsp70) and the 26 S proteasome in regulating apoptosis, although the precise nature of their involvement is not known. In the present study, control and Bcl-x(L)-overexpressing, interleukin-3-dependent FL5.12 cell lines were **treated** with the proteasome inhibitor N-benzoyloxycarbonyl (Z)-Leu-Leu-leucinal (MG132). Basal proteasome activity appeared to be approximately 30% lower in bcl-x(L) cells compared with control cells using a substrate for the chymotrypsin-like activity. However, no difference in proteasome activity was detected using substrates for the trypsin-like or peptidylglutamyl peptide-hydrolysing activities. In addition, protein levels of the 20 S proteasome beta-subunit, as determined by Western blot analyses, were similar in control and bcl-x(L) cells, leading to the conclusion that proteasome activities were the same in these two cell lines. At 24 h after **treatment** with 500 nM MG132, apoptosis in bcl-x(L) cells (22%) was less than that observed in control cells (34%). Concomitantly, caspase activity in control cells, as assessed by N-acetyl-l-aspartyl-l-glutamyl-l-valyl-l-aspartyl-7-amino-4-methylcou marin (Ac-DEVD-AMC), was twice that observed in bcl-x(L) cells. By 48 h after MG132 **treatment**, apoptosis and caspase activity in bcl-x(L) cells were similar to those observed in control cells at 24 h. Proteasome inhibition stimulated increases in hsp70 protein levels in control and bcl-x(L) cells by 12 h, although the maximal increases found in bcl-x(L) cells were less. Blocking this induction with hsp70 **antisense** oligonucleotides potentiated apoptosis after **treatment** with MG132. Inhibiting caspase activity with a broad-spectrum caspase inhibitor, t-butoxycarbonyl-Asp(OMe)-fluoromethyl ketone, prevented MG132-induced apoptosis. The more specific caspase-3 inhibitor, Ac-DEVD-aldehyde, afforded less protection, although both inhibitors completely inhibited Ac-DEVD-AMC cleavage. These data indicate that both hsp70 and Bcl-x(L) provide some protection against proteasome inhibitor-induced apoptosis.

4/3,AB/6 (Item 6 from file: 155)
DIALOG(R)File 155:MEDLINE(R)
(c) format only 2001 Dialog Corporation. All rts. reserv.

10453482 20050316 PMID: 10583267
Antisense-mediated suppression of **Bcl-2** highlights its pivotal role in failed apoptosis in B-cell chronic lymphocytic leukaemia.
Pepper C; Thomas A; Hoy T; Cotter F; Bentley P
Department of Haematology, Llandough Hospital, Penarth, South Glamorgan,

London.
British journal of [redacted]ematology (ENGLAND) Dec 199[redacted] 107 (3)
p611-5, ISSN 0007-1048 Journal Code: AXC
Languages: ENGLISH
Document type: Journal Article
Record type: Completed
Although advances have been made in the development of more effective **treatment** modalities, B-cell chronic lymphocytic leukaemia (B-CLL) remains incurable due to the development of drug resistance. Defective programmed cell death mechanisms rather than dysregulation of cell cycle appears to predominate in B-CLL and it is likely that a failure to initiate apoptosis contributes to chemoresistance. Most B-CLL cells contain high levels of the anti-apoptotic protein **Bcl-2** and high **Bcl-2** /Bax ratios have been associated with in vitro resistance to cytotoxic agents. In this study we evaluated the cellular responses to a **Bcl-2 antisense** oligonucleotide in terms of **Bcl-2** mRNA and protein expression and the induction of apoptosis. The **antisense** molecule induced a specific reduction in **Bcl-2** mRNA and protein expression over the 48 h culture period and was associated with increased apoptosis. The study indicates that **Bcl-2** protein is central to the mediation of resistance to apoptosis in B-CLL. Therefore **Bcl-2 antisense** oligonucleotides might be useful in the **treatment** of B-CLL.

4/3,AB/7 (Item 7 from file: 155)
DIALOG(R)File 155:MEDLINE(R)
(c) format only 2001 Dialog Corporation. All rts. reserv.

10437223 20071143 PMID: 10602518
Both FGF1 and bcl-x synthesis are necessary for the reduction of apoptosis in retinal pigmented epithelial cells by FGF2: role of the extracellular signal-regulated kinase 2.
Bryckaert M; Guillonneau X; Hecquet C; Courtois Y; Mascarelli F
INSERM U. 348, IFR Circulation, 75010 Paris, France.
Oncogene (ENGLAND) Dec 9 **1999**, 18 (52) p7584-93, ISSN 0950-9232 Journal Code: ONC
Languages: ENGLISH
Document type: Journal Article
Record type: Completed
Retinal pigmented epithelial (RPE) cells are of central importance in the maintenance of neural retinal function. Changes in the RPE cells associated with repair activities have been described as metaplasia, while RPE cell apoptosis is responsible for the development of a variety of retinal degenerations. We investigated the regulation of the anti-apoptotic properties of the fibroblast growth factors (FGF) 2 in serum-free cultures of RPE cells. In the absence of serum, confluent stationary RPE cells died by apoptosis via a caspase 3-dependent pathway. The addition of FGF2 greatly reduced apoptosis over a 7-day culture period. We demonstrated the involvement of an autocrine loop involving endogenous FGF1 in the mechanisms that govern FGF2-induced resistance to apoptosis by showing: (1) higher levels of apoptosis in cells **treated** with **antisense** FGF1 oligonucleotide or after neutralization of excreted FGF1; (2) the long-term activation of FGFR1 and of ERK2, (3) the inhibition of FGFR1 and ERK2 activation and an increase in apoptosis if excreted FGF1 was neutralized. FGF2 also increased the de novo synthesis and the production of Bcl-xl before the onset of apoptosis. Both inhibition of ERK2 activation, which decreased Bcl-xl synthesis, and downregulation of Bcl-x by **antisense** oligonucleotide **treatment** inhibited the survival-promoting activity of FGF2. Thus, FGF2-induced cell survival is a progressive adaptive phenomenon involving ERK2 activation by excreted FGF1 and ERK2-dependent Bcl-x production.

4/3,AB/8 (Item 8 from file: 155)

DIALOG(R)File 155:MEDLINE(R)
(c) format only 2001 Dialog Corporation. All rts. reserv.

10437185 20065090 PMID: 10597222
Key role of the cyclin-dependent kinase inhibitor p27kip1 for embryonal carcinoma cell survival and differentiation.
Baldassarre G; Barone MV; Belletti B; Sandomenico C; Bruni P; Spiezia S; Boccia A; Vento MT; Romano A; Pepe S; Fusco A; Viglietto G
Servizio Oncologia Sperimentale E, Istituto Nazionale Tumori, Napoli, Italy.
Oncogene (ENGLAND) Nov 4 **1999**, 18 (46) p6241-51, ISSN 0950-9232 Journal Code: ONC
Languages: ENGLISH
Document type: Journal Article
Record type: Completed
Hexamethylen-bisacetamide (HMBA) represents the prototype of a group of hybrid polar compounds, which induce differentiation in a variety of transformed cells including human embryonal carcinoma cells. Therefore, HMBA has been used in the differentiation therapy of cancer for patients with both hematological and solid malignancies. Upon HMBA **treatment**, the embryonal carcinoma cell line NTERA-2 clone D1 (NT2/D1) accumulates in G1 and undergoes terminal differentiation. Here we demonstrate that growth arrest and differentiation of NT2/D1 cells induced by HMBA involve increased expression of the cyclin-dependent kinase inhibitor p27, enhanced association of p27 with cyclin E/CDK2 complexes and suppression of kinase activity associated to cyclin E/CDK2 (but not to cyclin D3/CDK4). When HMBA differentiation was induced in the presence of p27 **antisense** oligonucleotides, NT2/D1 cells failed to arrest growth properly and, in parallel with the reduction of the anti-apoptotic **Bcl-2** gene expression, cells underwent massive programmed cell death. Conversely, constitutive expression of p27 into NT2/D1 cells induced a marked reduction in the growth potential of these cells and partially reproduced HMBA-induced modification of surface antigen expression (down-regulation of SSEA-3 expression and up-regulation of VINIS-53 expression). Expression of p21 induced growth arrest but not differentiation. Likewise, inhibition of CDK2 by transfection of a dominant negative CDK2 in NT2/D1 cells or **treatment** with the kinase inhibitor olomucine induced growth arrest but not differentiation. Therefore, we propose that p27 represents a crucial molecule in HMBA signaling that cannot be replaced by p21. Furthermore, the results obtained with CDK2 inhibitors demonstrate that the block of CDK2 activity is sufficient for growth arrest but not for cell differentiation and suggest that, at least in these cells, growth arrest and differentiation are regulated by two overlapping but different pathways.

4/3,AB/9 (Item 9 from file: 155)
DIALOG(R)File 155:MEDLINE(R)
(c) format only 2001 Dialog Corporation. All rts. reserv.

10434964 20069744 PMID: 10601320
Connective tissue growth factor induces apoptosis in human breast cancer cell line MCF-7.
Hishikawa K; Oemar BS; Tanner FC; Nakaki T; Luscher TF; Fujii T
Department of Pharmacology, Teikyo University School of Medicine, Tokyo 173-8605, Japan. hisikawa@med.teikyo-u.ac.jp
Journal of biological chemistry (UNITED STATES) Dec 24 **1999**, 274 (52) p37461-6, ISSN 0021-9258 Journal Code: HIV
Languages: ENGLISH
Document type: Journal Article
Record type: Completed
Connective tissue growth factor (CTGF) is a member of an emerging CCN gene family that is implicated in various diseases associated with fibro-proliferative disorder including scleroderma and atherosclerosis. The function of CTGF in human cancer is largely unknown. We now show that CTGF

induces apoptosis in the human breast cancer cell line MCF-7. CTGF mRNA was completely absent in MCF-7 but strongly induced by **treatment** with transforming growth factor beta (TGF-beta). TGF-beta by itself induced apoptosis in MCF-7, and this effect was reversed by co-**treatment** with CTGF **antisense** oligonucleotide. Overexpression of CTGF gene in transiently transfected MCF-7 cells significantly augmented apoptosis. Moreover, recombinant CTGF protein significantly enhanced apoptosis in MCF-7 cells as evaluated by DNA fragmentation, Tdt-mediated dUTP biotin nick end-labeling staining, flow cytometry analysis, and nuclear staining using Hoechst 33258. Finally, recombinant CTGF showed no effect on Bax protein expression but significantly reduced Bcl2 protein expression. Taken together, these results suggest that CTGF is a major inducer of apoptosis in the human breast cancer cell line MCF-7 and that TGF-beta-induced apoptosis in MCF-7 cells is mediated, in part, by CTGF.

4/3,AB/10 (Item 10 from file: 155)
DIALOG(R)File 155:MEDLINE(R)
(c) format only 2001 Dialog Corporation. All rts. reserv.

10433461 20036965 PMID: 10567678
BHRF1 **antisense** oligonucleotide inhibits anti-apoptosis of nasopharyngeal carcinoma cells.
Huang H; Pan X; Zhou J
Microbiology and Tumorbiology Center, Karolinska Institute, Stockholm S-17177, Sweden.
International journal of molecular medicine (GREECE) Dec **1999**, 4 (6) p649-53, ISSN 1107-3756 Journal Code: C8H
Languages: ENGLISH
Document type: Journal Article
Record type: Completed
Epstein-Barr virus gene BHRF1 has homology with proto-oncogene **bcl-2** , which can protect cells from apoptosis, thus, it may play important roles in oncogenesis and affect **treatment** of EBV-related cancers. We used BHRF1 **antisense** oligonucleotide to block its expression in nasopharyngeal carcinoma cell line, CNE2. The results showed that after blocking by BHRF1 **antisense** oligonucleotide, CNE2 cells had higher S-phase cell percentage, more susceptibility to radiation with weaker ability of proliferation, colony forming efficiency and tumor development in nude mice after radiation. Our results suggest that BHRF1 **antisense** oligonucleotide could inhibit BHRF1 anti-apoptotic ability, and may contribute to the **treatment** of EBV-related cancers.

4/3,AB/11 (Item 11 from file: 155)
DIALOG(R)File 155:MEDLINE(R)
(c) format only 2001 Dialog Corporation. All rts. reserv.

10429936 20072299 PMID: 10606283
Targeting **bcl-2** gene to delay androgen-independent progression and enhance chemosensitivity in prostate cancer using **antisense bcl-2** oligodeoxynucleotides.
Gleave ME; Miayake H; Goldie J; Nelson C; Tolcher A
The Prostate Centre, Vancouver General Hospital, British Columbia, Canada.
Urology (UNITED STATES) Dec **1999**, 54 (6A Suppl) p36-46, ISSN 0090-4295 Journal Code: WSY
Languages: ENGLISH
Document type: Journal Article
Record type: Completed
Bcl-2 expression is upregulated in prostate cancer cells after androgen withdrawal and is associated with the development of androgen independence and chemoresistance. Induction of apoptotic cell death after androgen ablation, or chemotherapy, may be enhanced through functional inhibition of **bcl-2**. In this report, we tested the

effects of **antisense** **bcl-2** oligodeoxynucleotides (ODN) with androgen ablation and taxane therapy on time to androgen-independent (Al) progression in the androgen-dependent Shionogi tumor model. **Treatment** of Shionogi tumor cells in vitro with 500 nmol/L **antisense** **bcl-2** ODN decreased **bcl-2** mRNA by 85%, compared with **treatment** with 500 nmol/L mismatch control ODN. Although **bcl-2** expression levels in Shionogi cells were not changed by docetaxel **treatment**, docetaxel **treatment** induced **bcl-2** phosphorylation. Consequently, the formation of **bcl-2** /Bax heterodimer formation was inhibited in a dose-dependent manner. **Treatment** of Shionogi tumors in vitro with either 500 nmol/L **antisense** **bcl-2** ODN or 10 nmol/L docetaxel alone did not induce apoptosis or reduce growth rates. However, combined **treatment** reduced the concentration that reduces cell viability by 50% (IC50) of docetaxel from 100 nmol/L to 10 nmol/L and induced characteristic apoptotic DNA laddering and cleavage of the poly(ADP-ribose)polymerase (PARP) protein. Adjuvant in vivo administration of **antisense** **bcl-2** ODN and polymeric micellar paclitaxel after castration resulted in a significant delay in time to Al recurrence when compared with administration of either agent alone. Furthermore, combined **treatment** of mice bearing Al recurrent Shionogi tumors with **antisense** **bcl-2** ODN and micellar paclitaxel synergistically induced tumor regression and growth inhibition when compared with **treatment** with either agent alone. These findings suggest that down-regulation of **bcl-2** by **antisense** ODN chemosensitizes Al Shionogi tumors to taxanes, over and above the effects of taxane-induced phosphorylation of **bcl-2**. **Antisense** **bcl-2** ODN combined with taxanes may be a novel approach to the **treatment** of both established and emerging Al disease.

4/3,AB/12 (Item 12 from file: 155)
DIALOG(R)File 155:MEDLINE(R)
(c) format only 2001 Dialog Corporation. All rts. reserv.

10426720 20065064 PMID: 10597196
p21Waf1/Cip1 acts in synergy with **bcl-2** to confer multidrug resistance in a camptothecin-selected human lung-cancer cell line.
Zhang Y; Fujita N; Tsuruo T
Institute of Molecular and Cellular Biosciences, University of Tokyo, Japan.
International journal of cancer. Journal international du cancer (UNITED STATES) Dec 10 **1999**, 83 (6) p790-7, ISSN 0020-7136
Journal Code: GQU
Languages: ENGLISH
Document type: Journal Article
Record type: Completed
The realization that chemotherapeutic agents induce apoptosis raises the concern that tumors resistant to chemotherapy are unable to initiate the apoptotic program. In the present study, we examined the apoptosis-resistance mechanism of a multidrug-resistant cell line, A549/CPT, which was established from the human lung-cancer cell line A549 by in vitro selection with gradually increased camptothecin (CPT) concentrations. We found that A549/CPT cells were resistant to anti-cancer drug-induced apoptosis in which caspase-3-like protease activity was attenuated remarkably, compared with parental A549 cells. We observed 2 mechanisms associated with apoptosis resistance in A549/CPT cells: over-expression of anti-apoptotic **bcl-2** and elevated expression of p21Waf1/Cip1. Transfection of either **bcl-2** or p21Waf1/Cip1 cDNA into parental A549 cells resulted in resistance to apoptosis. Furthermore, the co-**treatment** of p21Waf1/Cip1 and **bcl-2** anti-sense oligodeoxy-nucleotides restored drug susceptibility in A549/CPT cells more effectively than either one of them alone. These results indicate that co-induction of **bcl-2** and p21Waf1/Cip1 in A549/CPT cells may be involved in acquired drug resistance by inhibiting

caspase-mediated apoptosis. Agents aimed at preventing both **bcl-2** and p21Waf1/Cip1 expression may increase the efficiency of chemotherapy.

4/3,AB/13 (Item 13 from file: 155)
DIALOG(R)File 155:MEDLINE(R)
(c) format only 2001 Dialog Corporation. All rts. reserv.

10420438 20036552 PMID: 10567382
Inhibition of "tissue" transglutaminase increases cell survival by preventing apoptosis.
Oliverio S; Amendola A; Rodolfo C; Spinedi A; Piacentini M
Department of Biology, University of Rome "Tor Vergata," 00133 Rome, Italy.
Journal of biological chemistry (UNITED STATES) Nov 26 **1999**, 274 (48) p34123-8, ISSN 0021-9258 Journal Code: HIV
Languages: ENGLISH
Document type: Journal Article
Record type: Completed
Treatment of the human promonocytic cell line U937 with all-trans-retinoic acid (RA) commits these cells to apoptosis, which can be triggered by simply increasing intracellular calcium levels by the ionophore A23187. RA **treatment** of U937 cells is characterized by a decrease in **Bcl-2** and marked induction of "tissue" transglutaminase (tTG) gene expression. In this study, we show that the inhibition of tTG expression in U937 cells undergoing apoptosis prevents their death. In fact, U937 cell-derived clones transfected with the human tTG gene in the **antisense** orientation showed a pronounced decrease in apoptosis induced by several stimuli. These findings demonstrate that the Ca(2+)-dependent irreversible cross-linking of intracellular proteins catalyzed by tTG represents an important biochemical event in the gene-regulated cell death in monoblasts. In addition, our data indicate that the apoptotic program in promonocytic cells is strictly regulated by RA and that a key role is played by the free intracellular calcium concentration.

4/3,AB/14 (Item 14 from file: 155)
DIALOG(R)File 155:MEDLINE(R)
(c) format only 2001 Dialog Corporation. All rts. reserv.

10397934 99458856 PMID: 10527623
Involvement of cyclin-dependent kinase activities in CD437-induced apoptosis.
Hsu SL; Yin SC; Liu MC; Reichert U; Ho WL
Department of Education, Taichung Veterans General Hospital, Taichung, Taiwan. h2326@vghtc.vghtc.gov.tw
Experimental cell research (UNITED STATES) Nov 1 **1999**, 252 (2) p332-41, ISSN 0014-4827 Journal Code: EPB
Languages: ENGLISH
Document type: Journal Article
Record type: Completed
A novel synthetic retinoid, 6-[3-(1-adamantyl)-4-hydroxyphenyl]-2-naphthalene carboxylic acid (CD437), is a selective ligand of the RARgamma nuclear receptor. We examined the in vitro effects of CD437 and found that CD437 induces S phase arrest within 24 to 48 h, followed by cell death, in the p53-negative Hep3B and the p53-positive HepG2 human hepatoma cell lines. Based on observations of cellular and nuclear fragmentation, chromatin condensation, and DNA fragmentation, the CD437-mediated cell-killing effect appears to be due to apoptosis. On morphological examination, a number of CD437-**treated** cells were found to have increased 5- to 10-fold in size and persisted as single giant cells without cell division, while the remainder underwent nuclear division (multiple nuclei) but were unable to complete cytokinesis, and finally all died by apoptosis. In HepG2 cells

that possessed wild-type p53, CD437-induced S phase arrest and apoptosis were accompanied by up-regulation of cyclin A, cyclin B, p53, p21(CIP1/Waf1), Bad, and Bcl-Xs proteins and by a decrease in **Bcl-2** protein levels. In Hep3B cells, CD437-mediated S phase arrest and apoptosis were also associated with a concomitant up-regulation of cyclin A, cyclin B, Bad, and Bcl-Xs. However, Hep3B cells did not express p53 or **Bcl-2** messages. Olomoucine and roscovitine, the potent p34(cdc2) and CDK2 inhibitors, effectively blocked CD437-mediated cyclin A- and B-dependent kinase activation and prevented CD437-induced cell death. Furthermore, **antisense** oligonucleotide complementary to cyclin A and B mRNA significantly rescued CD437-induced apoptosis. These findings suggest that activation of cyclin A- and B-dependent kinases is a critical determinant of apoptotic death mediated by CD437. Copyright 1999 Academic Press.

4/3,AB/15 (Item 15 from file: 155)
DIALOG(R)File 155:MEDLINE(R)
(c) format only 2001 Dialog Corporation. All rts. reserv.

10393167 20015265 PMID: 10545916
Induction of endogenous Bcl-xS through the control of Bcl-x pre-mRNA splicing by **antisense** oligonucleotides.
Taylor JK; Zhang QQ; Wyatt JR; Dean NM
Department of Pharmacology, Isis Pharmaceuticals, Carlsbad, CA 92008, USA.
Nature biotechnology (UNITED STATES) Nov **1999**, 17 (11) p1097-100, ISSN 1087-0156 Journal Code: CQ3
Comment in Nat Biotechnol. 1999 Nov;17(11) 1064-5
Languages: ENGLISH
Document type: Journal Article
Record type: Completed
Resistance to apoptosis, which plays an important role in tumors that are refractory to chemotherapy, is regulated by the ratio of antiapoptotic to proapoptotic proteins. By manipulating levels of these proteins, cells can become sensitized to undergo apoptosis in response to chemotherapeutic agents. Alternative splicing of the bcl-x gene gives rise to two proteins with antagonistic functions: Bcl-xL, a well-characterized antiapoptotic protein, and Bcl-xS, a proapoptotic protein. We show here that altering the ratio of Bcl-xL to Bcl-xS in the cell using an **antisense** oligonucleotide permitted cells to be sensitized to undergo apoptosis in response to ultraviolet B radiation and chemotherapeutic drug **treatment** . These results demonstrate the ability of a chemically modified oligonucleotide to alter splice site selection in an endogenous gene and illustrate a powerful tool to regulate cell survival.

4/3,AB/16 (Item 16 from file: 155)
DIALOG(R)File 155:MEDLINE(R)
(c) format only 2001 Dialog Corporation. All rts. reserv.

10386353 20012565 PMID: 10547074
Role of CD14 expression in the differentiation-apoptosis switch in human monocytic leukemia cells **treated** with 1alpha,25-dihydroxyvitamin D3 or dexamethasone in the presence of transforming growth factor beta1.
Kanatani Y; Kasukabe T; Okabe-Kado J; Yamamoto-Yamaguchi Y; Nagata N; Motoyoshi K; Honma Y
Saitama Cancer Center Research Institute, Ina, Japan.
Cell growth & differentiation (UNITED STATES) Oct **1999**, 10 (10) p705-12, ISSN 1044-9523 Journal Code: AYH
Languages: ENGLISH
Document type: Journal Article
Record type: Completed
Transforming growth factor beta (TGF-beta) enhanced the growth-inhibitory activities of dexamethasone (Dex) and 1alpha,25-dihydroxyvitamin D3 (VD3)

on human monocytoid leukemia U937 cells. TGF-beta and VD3 synergistically increased the expression of differentiation-associated markers such as the CD11b and CD14 antigens, whereas TGF-beta and Dex did not. On the other hand, TGF-beta and Dex synergistically increased the number of Apo2.7-positive cells, which represents the early stage of apoptosis, whereas TGF-beta and VD3 did not, suggesting that TGF-beta enhanced apoptosis with Dex and enhanced monocytic differentiation with VD3. In the presence of TGF-beta, the retinoblastoma susceptibility gene product, pRb, was synergistically dephosphorylated by Dex as well as VD3. TGF similarly enhanced the expression of the p21Waf1 gene in U937 cells **treated** with Dex and VD3. TGF-beta dose-dependently increased the expression of **Bcl-2** and Bad and decreased the expression of Bcl-X(L) in U937 cells. Dex enhanced the down-regulation of Bcl-X(L) expression in TGF-beta-**treated** cells, whereas VD3 blocked this down-regulation of Bcl-X(L). However, the down-regulation of Bcl-X(L) by **treatment** with the **antisense** oligomer did not affect the apoptosis or differentiation of U937 cells. The apoptosis of CD14-positive cells was suppressed in the VD3 plus TGF-beta-**treated** cultures. These results suggest that the expression of CD14 is involved in the survival of differentiated cells.

4/3,AB/17 (Item 17 from file: 155)
DIALOG(R)File 155:MEDLINE(R)
(c) format only 2001 Dialog Corporation. All rts. reserv.

10378277 99458294 PMID: 10530711

Antisense therapy of hematologic malignancies.

Cotter FE
Department of Experimental Hematology, St. Bartholomew Royal London School of Medicine, UK.

Seminars in hematology (UNITED STATES) Oct **1999**, 36 (4 Suppl 6) p9-14, ISSN 0037-1963 Journal Code: UN9
Languages: ENGLISH
Document type: Journal Article; Review; Review, Tutorial
Record type: Completed

Many tumor cells are inherently resistant to curative **treatment** due to an altered pattern of gene expression. It is an attractive and logical proposition to use this difference within the lymphoma cell to eradicate the malignant process. One such new therapeutic approach based on the "silencing" of genes involved in the prevention of apoptosis is **Bcl-2 antisense** oligonucleotide (AO) therapy. In the field of lymphoma, obvious targets included follicular lymphoma with the t(15;18) translocation, which results in deregulated expression of the **Bcl-2** gene, chemoresistance, and subsequent protection against lymphoma cell death. Targeting the initiating codon of the **Bcl-2** gene decreases both cell viability and **Bcl-2** protein expression in lymphoma and leukemia cell lines that overexpress **Bcl-2**. Preclinical toxicity studies using a **Bcl-2** AO G3139 (Genta, San Diego, CA) show good tolerance at a dose of 10 mg/kg, which is considerably higher than the dose required for good antilymphoma efficacy. In a phase I clinical study, G3139 was well tolerated with minimal toxicity in a dose escalation up to 147.2 mg/m2/d. Evidence of efficacy includes a responder with stage IVB follicular lymphoma who achieved complete clinical and radiologic response that has lasted more than 2 years. The main dose-limiting toxicity has been reversible thrombocytopenia related to the thioate backbone. Other **antisense** reagents are also in development to combat non-Hodgkin's lymphoma (NHL). These include oligonucleotides that target the messages of the Bcl-X(L) and protein kinase-Calpha (PKCalpha) genes. AOs may also have an application in tumors expressing mutant p53. AOs against MDM2 genes have shown the ability to restore wild-type p53 expression, suggesting that as oncogenic pathways are unraveled, normal cell growth and death patterns may be restored by molecular manipulation. Downregulation of antiapoptosis by AOs in the human setting has low toxicity and antilymphoma activity in cases in which conventional chemotherapy has failed. In the future, **antisense** therapy followed by

chemotherapy may overcome chemoresistance to provide effective therapy for a range of malignancies.

4/3,AB/18 (Item 18 from file: 155)
DIALOG(R)File 155:MEDLINE(R)
(c) format only 2001 Dialog Corporation. All rts. reserv.

10373391 99320989 PMID: 10395066
Growth arrest and induction of apoptosis in breast cancer cells by **antisense** depletion of protein kinase A-RI alpha subunit: p53-independent mechanism of action.
Srivastava RK; Srivastava AR; Seth P; Agrawal S; Cho-Chung YS
Cellular Biochemistry Section, Laboratory of Tumor Immunology and Biology, National Cancer Institute, National Institutes of Health, Bethesda, Maryland 20892-1750, USA.
Molecular and cellular biochemistry (NETHERLANDS) May **1999**, 195 (1-2) p25-36, ISSN 0300-8177 Journal Code: NGU
Languages: ENGLISH
Document type: Journal Article
Record type: Completed
The enhanced expression of the RI alpha subunit of cyclic AMP-dependent protein kinase type 1 (PKA-1) has been correlated with cancer cell growth. We have investigated the effects of sequence-specific inhibition of RI alpha gene expression on the growth of MCF-7 human breast cancer cells. We report that RI alpha **antisense treatment** results in a reduction in RI alpha expression at both mRNA and protein levels and inhibition of cell growth. The growth inhibition was accompanied by changes in cell morphology, cleavage of poly(ADP-ribose) polymerase (PARP) and appearance of apoptotic nuclei. In addition, **bcl-2** protein level was reduced and p53 expression increased in growth arrested cells. Interestingly, RI alpha **antisense** inhibited cell viability and induced apoptosis in the absence of p53, suggesting that these actions of RI alpha **antisense** are exerted independent of p53. In contrast, two- and four-base mismatched control oligonucleotides had no effect on either cell growth or morphology. These results demonstrate that the RI alpha **antisense**, which efficiently depletes the growth stimulatory molecule RI alpha, induces cell differentiation and apoptosis, providing a new approach to combat breast cancer cell growth.

4/3,AB/19 (Item 19 from file: 155)
DIALOG(R)File 155:MEDLINE(R)
(c) format only 2001 Dialog Corporation. All rts. reserv.

10365729 99428520 PMID: 10497210
The endoplasmic reticulum chaperone glycoprotein GRP94 with Ca(2+)-binding and antiapoptotic properties is a novel proteolytic target of calpain during etoposide-induced apoptosis.
Reddy RK; Lu J; Lee AS
Department of Biochemistry, USC/Norris Comprehensive Cancer Center, University of Southern California School of Medicine, Los Angeles, California 90033, USA.
Journal of biological chemistry (UNITED STATES) Oct 1 **1999**, 274 (40) p28476-83, ISSN 0021-9258 Journal Code: HIV
Contract/Grant No.: CA27607, CA, NCI; EY03040, EY, NEI
Languages: ENGLISH
Document type: Journal Article
Record type: Completed
GRP94 is a 94-kDa chaperone glycoprotein with Ca(2+)-binding properties. We report here that during apoptosis induced by the topoisomerase II inhibitor etoposide, a fraction of GRP94 associated with the endoplasmic reticulum membrane undergoes specific proteolytic cleavage, coinciding with the activation of the caspase CPP32 and initiation of DNA fragmentation. In vivo, inhibitors of caspases able to block etoposide-induced apoptosis can

only partially protect GRP94 from proteolytic cleavage, whereas complete inhibition is observed with calpain inhibitor I but not with the proteasome inhibitor. In vitro, GRP94 is not a substrate for CPP32; rather, it can be completely cleaved by calpain, a Ca(2+)-regulated protease. The cleavage of GRP94 by calpain is Ca(2+)-dependent and generates a discrete polypeptide of 80 kDa. In contrast, calpain has no effect on other stress proteins such as GRP78 or HSP70. Further, immunohistochemical staining reveals specific co-localization of GRP94 with calpain in the perinuclear region following etoposide **treatment**. We further showed that reduction of GRP94 by **antisense** decreased cell viability in etoposide-**treated** Jurkat cells. Our studies provide new evidence that the cytoprotective GRP94, as in the case of the antiapoptotic protein **Bcl-2**, can be targets of proteolytic cleavage themselves during the apoptotic process.

4/3,AB/20 (Item 20 from file: 155)
DIALOG(R)File 155:MEDLINE(R)
(c) format only 2001 Dialog Corporation. All rts. reserv.

10364515 99452899 PMID: 10521802
Protein-kinase-C iso-enzymes support DNA synthesis and cell survival in colorectal-tumor cells.
Hochegger K; Partik G; Schorkhuber M; Marian B
Institute of Tumor Biology/Cancer Research, University of Vienna, Vienna, Austria.
International journal of cancer. Journal international du cancer (UNITED STATES) Nov 26 **1999**, 83 (5) p650-6, ISSN 0020-7136
Journal Code: GQU
Languages: ENGLISH
Document type: Journal Article
Record type: Completed
Protein-kinase-C signalling has been blocked in colorectal tumor cells by kinase inhibitors, by TPA down-regulation or by exposure to anti-sense oligonucleotides. This resulted in growth inhibition in all cell lines used. The kinase inhibitors H7 and calphostin induced apoptosis, demonstrated by the appearance of cells with characteristically condensed chromatin and the induction of stand-breaks in the DNA. A cell-death-inducing concentration of 15 microgram/ml H7 down-regulated the **bcl-2** levels after 9 hr, while bak levels were not affected. Go6976,-an inhibitor of Ca(++)-dependent PKC iso-enzymes, was not active in growth inhibition or induction of apoptosis. Analysis of DNA synthesis in inhibitor-**treated** cultures indicated that H7 caused strong inhibition in all cell lines, while the more specific inhibitor calphostin was effective only in VACO235 adenoma cells. When down-regulation by TPA or anti-sense oligonucleotides was used to block PKC, effects on cell numbers were smaller and delayed. However, induction of apoptosis was significantly increased in SW480 carcinoma cells 4 days after exposure to anti-epsilon and anti-zeta oligonucleotides in SW480 and T84 carcinoma cells. Apoptosis was preceeded by loss of PKC protein and of **bcl-2** from day 1 after addition of the oligonucleotides. In VACO235 adenoma cells, no induction of apoptosis could be observed when anti-epsilon and anti-zeta oligonucleotides were used. On the other hand, the adenoma cells were more responsive to anti-alpha and anti-beta oligonucleotides, which strongly inhibited DNA-synthesis 3 days after addition to the culture medium. Our results indicate that the Ca(++)-dependent PKCs alpha and beta are involved in proliferation signals, while the Ca(++)-independent PKCs epsilon and zeta are involved in survival pathways of colorectal tumor cells. Copyright 1999 Wiley-Liss, Inc.

4/3,AB/21 (Item 21 from file: 155)
DIALOG(R)File 155:MEDLINE(R)
(c) format only 2001 Dialog Corporation. All rts. reserv.

10360822 20019590 PMID: 10554162

Synthesis of **Bcl-2** in response to anthracycline **treatment** may contribute to an apoptosis-resistant phenotype in leukemic cell lines.

Durrieu F; Belaud-Rotureau MA; Lacombe F; Dumain P; Reiffers J; Boisseau MR; Bernard P; Belloc F

Laboratoire Universitaire d'Hematologie, Universite Victor Segalen, Bordeaux, France.

Cytometry (UNITED STATES) Jun 1 **1999**, 36 (2) p140-9, ISSN 0196-4763 Journal Code: D92

Languages: ENGLISH

Document type: Journal Article

Record type: Completed

BACKGROUND: Some forms of chemoresistance in leukemia may start from failure of tumour cells to successfully undergo apoptosis and **Bcl-2** may play a role in this defect. Therefore, we evaluated the **Bcl-2** content and synthesis in relation with the apoptotic potential in leukemic cell lines after anthracycline **treatment**. METHODS: U937, HL60, and K562 cells and their drug resistant (DR) variants were **treated** with varying concentrations of Idarubicin (IDA). Apoptosis was evaluated by fluorescence microscopy after acridine orange staining. **Bcl-2** and Bax content were evaluated either by flow cytometry after indirect immunolabelling or by Western blot. RESULTS: High **Bcl-2** contents were not related to a poor ability to undergo apoptosis in U937, HL60, K562 and their DR variants. IDA induced a concentration-dependent increase in **Bcl-2** content in all cell lines as long as they do not perform apoptosis. Enhanced **Bcl-2** expression was inhibited by cycloheximide, actinomycin D, or **antisense** oligonucleotide directed against **bcl-2** mRNA. **Bcl-2** expression was also increased in the resistant U937 variant after serum deprivation or C2-ceramide **treatment** . The synthesis of **Bcl-2** led to an increased **Bcl-2**/Bax ratio solely in the cells with an apoptosis-resistance phenotype. CONCLUSIONS: These data suggest that exposure to IDA induces **Bcl-2** expression in leukemic cell lines, and that this mechanism could contribute to apoptosis resistance and participate in the acquisition of chemoresistance. They also confirm that the evolution of the **Bcl-2** /Bax ratio reflects apoptotic ability better than the steady state level of **Bcl-2** expression.

4/3,AB/22 (Item 22 from file: 155)

DIALOG(R)File 155:MEDLINE(R)

(c) format only 2001 Dialog Corporation. All rts. reserv.

10357930 20005750 PMID: 10537358

Progression to androgen independence is delayed by adjuvant **treatment** with **antisense Bcl-2** oligodeoxynucleotides after castration in the LNCaP prostate tumor model.

Gleave M; Tolcher A; Miyake H; Nelson C; Brown B; Beraldi E; Goldie J

Department of Cancer Endocrinology, British Columbia Cancer Agency, Vancouver, Canada.

Clinical cancer research (UNITED STATES) Oct **1999**, 5 (10) p2891-8, ISSN 1078-0432 Journal Code: C2H

Languages: ENGLISH

Document type: Journal Article

Record type: Completed

Bcl-2 has emerged as a critical regulator of apoptosis in a variety of cell systems and is up-regulated during progression to androgen independence in prostate cancer cells. The objectives of this study were to characterize changes in **Bcl-2** after androgen withdrawal and during progression to androgen independence in the human prostate LNCaP tumor model and determine whether adjuvant use of **antisense Bcl-2** oligodeoxynucleotides (ODNs) with androgen ablation delays progression to androgen independence. **Bcl-2** expression in LNCaP cells is down-regulated to undetectable levels by androgen in vitro and

up-regulated after castration in vivo. **Antisense Bcl-2** ODN **treatment** reduced [redacted]aP cell **Bcl-2** messenger RNA and protein levels by >90% in a sequence-specific and dose-dependent manner at concentrations >50 nM. **Bcl-2** mRNA levels returned to pretreatment levels by 48 h after discontinuing **treatment**. Athymic male mice bearing SQ LNCaP tumors were castrated and injected i.p. with 12.5 mg/kg/day with two-base mismatch ODN control, reverse polarity ODN control, or **antisense Bcl-2** ODN. Tumor volume in control mice gradually increased 5-fold (range, 3-6) by 12 weeks after castration compared to a 10-50% decrease in precastrate tumor volume in mice **treated** with **antisense Bcl-2** ODN. Changes in serum PSA paralleled changes in tumor volume, increasing 4-fold faster above nadir in controls than in mice **treated** with **antisense Bcl-2** ODN. After decreasing 70% by 1 week after castration, PSA increased 1.6-fold above precastrate levels by 11 weeks in controls while staying 30% below precastrate levels in **antisense-treated** mice. In a second group of experiments, LNCaP tumor growth and serum PSA levels were 90% lower ($P<0.01$) in mice **treated** with **antisense Bcl-2** ODN compared with mismatch or reverse polarity ODN controls. These results support the hypothesis that **Bcl-2** helps mediate progression to androgen independence and is an appropriate target for **antisense** therapy.

4/3,AB/23 (Item 23 from file: 155)
DIALOG(R)File 155:MEDLINE(R)
(c) format only 2001 Dialog Corporation. All rts. reserv.

10337386 99228212 PMID: 10213224

Cooperative inhibitory effect of novel mixed backbone oligonucleotide targeting protein kinase A in combination with docetaxel and anti-epidermal growth factor-receptor antibody on human breast cancer cell growth.

Tortora G; Caputo R; Pomatico G; Pepe S; Bianco AR; Agrawal S; Mendelsohn J; Ciardiello F

Cattedra di Oncologia Medica, Dipartimento di Endocrinologia e Oncologia Molecolare e Clinica, Universita Federico II, Napoli, Italy.

Clinical cancer research (UNITED STATES) Apr **1999**, 5 (4) p875-81, ISSN 1078-0432 Journal Code: C2H

Languages: ENGLISH

Document type: Journal Article

Record type: Completed

Type I protein kinase A (PKAI) is overexpressed in the majority of human tumors and plays a relevant role in neoplastic transformation, conveying mitogenic signals of different growth factors and oncogenes. Inhibition of PKAI by **antisense** oligonucleotides targeting its RIalpha regulatory subunit results in cancer cell growth inhibition in vitro and in vivo. We have recently shown that a mixed backbone oligonucleotide targeting RIalpha can cooperatively inhibit human cancer cell growth when combined with selected cytotoxic drugs. In the present study, we have used HYB 165, a novel DNA/RNA hybrid mixed backbone oligonucleotide that exhibits improved pharmacokinetic and bioavailability properties in vivo and is presently undergoing Phase I trials. We have shown that HYB 165 exhibits a dose-dependent inhibitory effect on ZR-75-1 cells and a cooperative activity with docetaxel, a cytotoxic drug active in breast cancer. The antiproliferative activity is accompanied by increased apoptosis, as compared with each single agent. On the basis of our previous demonstration of a structural and functional relation between PKAI and epidermal growth factor receptor, we have performed a double blockade of these pathways using HYB 165 in combination with monoclonal antibody (MAb) C225, an anti-epidermal growth factor receptor chimeric MAb. The two compounds determined a cooperative growth inhibitory effect on ZR-75-1 cells and increased apoptosis. To study whether different biological agents and cytotoxic drugs can interact together, low doses of HYB 165, MAb C225, and docetaxel were combined causing an even greater cooperative effect toward growth inhibition. Finally, we have demonstrated that each single agent is

able to induce **bcl-2** phosphorylation and that the three agents, used in combination at suboptimal doses, determine a greater degree of **bcl-2** phosphorylation and cause apoptosis of the majority of ZR-75-1 cells. These findings provide the basis for a novel strategy of **treatment** of breast cancer patients because both HYB 165 and MAb C225 are presently under clinical evaluation.

4/3,AB/24 (Item 24 from file: 155)
DIALOG(R)File 155:MEDLINE(R)
(c) format only 2001 Dialog Corporation. All rts. reserv.

10333805 99240640 PMID: 10222151
Selective activation of Ha-ras(val12) oncogene increases susceptibilityof NIH/3T3 cells to TNF-alpha.
Chang MY; Won SJ; Yang BC; Jan MS; Liu HS
College of Medicine, National Cheng Kung University, Tainan, Taiwan, Republic of China.
Experimental cell research (UNITED STATES) May 1 **1999**, 248 (2) p589-98, ISSN 0014-4827 Journal Code: EPB
Languages: ENGLISH
Document type: Journal Article
Record type: Completed
This is the first report demonstrating that NIH/3T3 fibroblasts utilize the Raf-1/MAPK pathway to sensitize themselves to tumor necrosis factor-alpha (TNF-alpha) cytotoxicity under Ha-rasVal12 oncogene-overexpressed conditions. This paper clearly shows that the sensitivity of NIH/3T3 cells to TNF-alpha cytotoxicity positively correlated with the expression level of activated Ha-ras transgene, which was manipulated either positively by isopropyl-beta-d-thiogalactoside (IPTG) induction or negatively by a **ribozyme** or a dominant negative Ras suppression. Further analysis revealed that after TNF-alpha **treatment**, Ha-ras-overexpressed transformants underwent apoptosis. Overexpression of dominant negative Raf-1, Rac1, or RhoA in the Ha-ras transformants clarified that among these factors, only dominant negative Raf-1 could reverse the cell sensitivity to TNF-alpha, indicating that Raf-1, as a proapoptotic factor, indeed participates in TNF-alpha cytotoxicity. The anti-apoptotic roles of **Bcl-2** and PI(3) kinase are also demonstrated by the Ha-ras transformants which became more resistant to TNF-alpha while overexpressing **Bcl-2** or the activated p110 catalytic subunit. The analyses of the cell cycle and nuclear transcription factor activities revealed that TNF-alpha **treatment** caused the Ha-ras overexpressed transformants to shift from S to G0/G1 phase and increased the responses of AP-1, c-fos, and c-myc. Taken together, we suggest that the possible action of Ha-ras overexpression to sensitize TNF-alpha-**treated** fibroblasts is predominantly through the Ras/Raf-1/MAPK pathway to increase the responses of AP-1, c-fos, and c-myc, which are possibly involved in the aberration of cell cycle machinery, and subsequently to turn on the death program. Copyright 1999 Academic Press.

4/3,AB/25 (Item 25 from file: 155)
DIALOG(R)File 155:MEDLINE(R)
(c) format only 2001 Dialog Corporation. All rts. reserv.

10333472 99244198 PMID: 10229192
Tiam1 is involved in the regulation of bufalin-induced apoptosis in human leukemia cells.
Kawazoe N; Watabe M; Masuda Y; Nakajo S; Nakaya K
Laboratory of Biological Chemistry, School of Pharmaceutical Sciences, Showa University, Tokyo, Japan.
Oncogene (ENGLAND) Apr 15 **1999**, 18 (15) p2413-21, ISSN 0950-9232 Journal Code: ONC
Languages: ENGLISH
Document type: Journal Article

Record type: Completed
Bufalin, a component of the Chinese medicine chan'su, induces apoptosis in various lines of human tumor cells, such as leukemia HL60 and U937 cells, by altering the expression of apoptosis-related genes, for example, **bcl-2** and c-myc. In this study, we characterized a gene that is involved in bufalin-induced apoptosis by the differential display (DD) technique. The partial nucleotide sequence of one of the differentially expressed clones obtained after **treatment** with bufalin was identical to that of the human gene for Tiam1. When U937 cells were **treated** with 10(-7) M bufalin, expression of both Tiam1 mRNA and the protein was induced 1 h after the start of the **treatment**. The increase of Tiam1 mRNA was transient but the level of Tiam1 protein continued to increase at least for 6 h. In addition, the activities of Rac1 and p21-activated kinase (PAK) were also stimulated by bufalin **treatment**. To evaluate the role of Tiam1 in the apoptotic process, we examined the effects of the expression of sense and **antisense** RNA for Tiam1 in U937 cells. Apoptosis was strongly induced by bufalin in cells that expressed sense RNA for Tiam1 as compared to apoptosis in control cells **treated** with bufalin only. Cells expressing **antisense** RNA for Tiam1 were significantly more resistant than the control bufalin-**treated** cells to induction of DNA fragmentation in response to bufalin. Moreover, sense transformants had elevated activities of PAK and c-Jun NH2-terminal kinase (JNK). These results suggest that Tiam1 might play a critical role in bufalin-induced apoptosis through the activation of Rac1, PAK, and JNK pathway.

4/3,AB/26 (Item 26 from file: 155)
DIALOG(R)File 155:MEDLINE(R)
(c) format only 2001 Dialog Corporation. All rts. reserv.

10331813 99185070 PMID: 10085086
Tumor necrosis factor induces **Bcl-2** and Bcl-x expression through NFkappaB activation in primary hippocampal neurons.
Tamatani M; Che YH; Matsuzaki H; Ogawa S; Okado H; Miyake S; Mizuno T; Tohyama M
Department of Anatomy and Neuroscience, Osaka University Medical School, 2-2 Yamadaoka, Suita, Osaka 565-0872, Japan. tama@anat2.med.osaka-u.ac.jp
Journal of biological chemistry (UNITED STATES) Mar 26 **1999**, 274 (13) p8531-8, ISSN 0021-9258 Journal Code: HIV
Languages: ENGLISH
Document type: Journal Article
Record type: Completed
Emerging data indicate that tumor necrosis factor (TNF) exerts a neuroprotective effect in response to brain injury. Here we examined the mechanism of TNF in preventing neuronal death in primary hippocampal neurons. TNF protected neurons against hypoxia- or nitric oxide-induced injury, with an increase in the anti-apoptotic proteins **Bcl-2** and Bcl-x as determined by Western blot and reverse transcriptase-polymerase chain reaction analysis. **Treatment** of neurons with an **antisense** oligonucleotide to **bcl-2** mRNA or that to bcl-x mRNA blocked the up-regulation of **Bcl-2** or Bcl-x expression, respectively, and partially inhibited the neuroprotective effect induced by TNF. Moreover, adenovirus-mediated overexpression of **Bcl-2** significantly inhibited hypoxia- or nitric oxide-induced neuronal death. To examine the possible involvement of a transcription factor, NFkappaB, in the regulation of **Bcl-2** and Bcl-x expression in TNF-**treated** neurons, an adenoviral vector capable of expressing a mutated form of IkappaB was used to infect neurons prior to TNF **treatment**. Expression of the mutant NFkappaB completely inhibited NFkappaB DNA binding activity and inhibited both TNF-induced up-regulation of **Bcl-2** and Bcl-x expression and neuroprotective effect. These findings indicate that induction of **Bcl-2** and Bcl-x expression through NFkappaB activation is involved in the neuroprotective action of TNF against hypoxia- or nitric oxide-induced injury.

4/3,AB/27 (Item 27 from file: 155)
DIALOG(R)File 155:MEDLINE(R)
(c) format only 2001 Dialog Corporation. All rts. reserv.

10312443 98421812 PMID: 9751192
Involvement of **Bcl-2** family and caspase-3-like protease in NO-mediated neuronal apoptosis.
Tamatani M; Ogawa S; Niitsu Y; Tohyama M
Department of Anatomy and Neuroscience, Osaka University Medical School, Suita, Japan.
Journal of neurochemistry (UNITED STATES) Oct **1998**, 71 (4) p1588-96, ISSN 0022-3042 Journal Code: JAV
Languages: ENGLISH
Document type: Journal Article
Record type: Completed
To clarify mechanisms of neuronal death in the postischemic brain, we examined whether astrocytes exposed to hypoxia/reoxygenation exert a neurotoxic effect, using a coculture system. Neurons cocultured with astrocytes subjected to hypoxia/reoxygenation underwent apoptotic cell death, the effect enhanced by a combination of interleukin-1beta with hypoxia. The synergistic neurotoxic activity of hypoxia and interleukin-1beta was dependent on de novo expression of inducible nitric oxide synthase (iNOS) and on nitric oxide (NO) production in astrocytes. Further analysis to determine the neurotoxic mechanism revealed decreased **Bcl-2** and increased Bax expression together with caspase-3 activation in cortical neurons cocultured with NO-producing astrocytes. Inhibition of NO production in astrocytes by N(G)-monomethyl-L-arginine, an inhibitor of NOS, significantly inhibited neuronal death together with changes in **Bcl-2** and Bax protein levels and in caspase-3-like activity. Moreover, **treatment** of neurons with a bax **antisense** oligonucleotide inhibited the caspase-3-like activation and neuronal death induced by an NO donor, sodium nitroprusside. These data suggest that NO produced by astrocytes after hypoxic insult induces apoptotic death of neurons through mechanisms involving the caspase-3 activation after down-regulation of **Bcl-2** and up-regulation of Bax protein levels.

4/3,AB/28 (Item 28 from file: 155)
DIALOG(R)File 155:MEDLINE(R)
(c) format only 2001 Dialog Corporation. All rts. reserv.

10306477 98251288 PMID: 9589383
The intracellular domain of p55 tumor necrosis factor receptor induces apoptosis which requires different caspases in naive and neuronal PC12 cells.
Haviv R; Stein R
Department of Neurobiochemistry, George S. Wise Faculty of Life Sciences, Tel-Aviv University, Ramat Aviv, Israel.
Journal of neuroscience research (UNITED STATES) May 15 **1998**, 52 (4) p380-9, ISSN 0360-4012 Journal Code: KAC
Languages: ENGLISH
Document type: Journal Article
Record type: Completed
Apoptosis is induced in cells via distinct pathways, which may differ according to various stimuli and different cell types. One apoptotic stimulus is the activation of receptors such as the p55 tumor necrosis factor (TNF) receptor. These receptors transduce their apoptotic signals via a cytoplasmic region termed the death domain. Here we investigated the apoptotic pathway induced by overexpression of the intracellular domain of p55 TNF receptor (p55-IC) in a neuronal model system consisting of PC12 cells. Using the tetracycline-regulated transactivator system, which allows controlled gene expression, we show that overexpression of p55-IC induces

apoptosis in both naive and neuronal PC12 cells. The apoptosis-inducing effect of p55-IC is blocked by the expression of **bcl-2**, suggesting that p55-IC induces apoptosis in PC12 cells via a pathway controlled by **bcl-2** . The need for caspases in the p55-IC-induced cell death effect in naive and neuronal PC12 cells was studied by examining the effects of broad-spectrum and specific inhibitors of caspases as well as expression of **antisense** caspase-2 RNA. The broad-spectrum caspase inhibitor benzyloxycarbonyl-Val-Ala-Asp-fluoromethyl -ketone blocked p55-IC-induced cell death in both naive and neuronal cells, suggesting that caspases are needed for this process in both cell types. Caspase-1-like proteases are most probably not involved in the process since neither expression of crmA nor **treatment** with the caspase-1-specific peptide inhibitor Ac-Try-Val-Ala-Asp-aldehyde had any protective effect. Interestingly, expression of **antisense** caspase-2 RNA blocked the p55-IC-induced cell death in naive but not in neuronal PC12 cells, whereas the caspase-3-like specific inhibitor Ac-Asp-Glu-Val-Asp-aldehyde partially inhibited this death in neuronal but not in naive cells. These results suggest that the apoptosis-inducing effect of p55-IC requires different caspases in naive and neuronal PC12 cells.

4/3,AB/29 (Item 29 from file: 155)
DIALOG(R)File 155:MEDLINE(R)
(c) format only 2001 Dialog Corporation. All rts. reserv.

10290083 97376497 PMID: 9232607

Enhanced expression of **bcl-2** following **antisense** oligonucleotide mediated growth factor deprivation.

Rubenstein M; Chou P; Mirochnik Y; Guinan P
Hektoen Institute for Medical Research, Department of Urology, Rush Presbyterian St Lukes Medical Center, Chicago, Illinois 60612, USA.

Medical oncology (ENGLAND) Mar **1997**, 14 (1) p23-9, ISSN 1357-0560 Journal Code: B3A
Languages: ENGLISH
Document type: Journal Article
Record type: Completed

Although the role of **bcl-2** in apoptosis has been described, its involvement in prostate cancer (CAP) progression is less well understood, but thought to be involved with the transition of CAP from androgen-sensitivity to androgen-independence, where its expression is augmented following androgen ablation. For **treating** these recurrent androgen-independent tumors, following hormone **treatment** failure, a new tier of therapy based upon growth factor deprivation has been suggested, implemented by **antisense** oligonucleotides (oligos) directed against mRNA encoding a critical growth regulatory autocrine loop (comprised of transforming growth factor-alpha (TGF-alpha) and its binding site, the epidermal growth factor receptor (EGFR). To determine whether oligo-induced growth factor deprivation therapy similarly enhanced expression of **bcl-2** (as follows androgen deprivation) human prostate cancer derived PC-3 cells were **treated** in vitro with oligos directed against TGF-alpha (MR-1) and/or EGFR (MR-2). After 5 days of **treatment** cells were immunochemically stained for human **bcl-2**. In similar experiments, cells were **treated** for 3 days prior to extraction of proteins, Western blot analysis, photography and computer evaluation of protein density by SigmaScan software. Immunostained cells **treated** with oligos directed against mRNA encoding TGF-alpha (MR-1) either alone or in combination with that directed against EGFR (MR-2) had increased **bcl-2** expression (+3 to +5). In addition, the intensity of Western blots scanned for **bcl-2** expression were 19%, 32% and 30% greater in cells **treated** with oligos directed against TGF-alpha, EGFR or their combination, respectively. We conclude that enhanced **bcl-2** expression followed **antisense** oligo induced growth factor deprivation. This result is similar to that found upon androgen deprivation therapy, and also demonstrates additional

biologic activity of this new class of molecular therapeutic agents.

4/3,AB/30 (Item 30 from file: 155)
DIALOG(R)File 155:MEDLINE(R)
(c) format only 2001 Dialog Corporation. All rts. reserv.

10277745 99316964 PMID: 10390076
Antisense strategy in hematological malignancies.
Warzocha K
Department of Hematology, Medical University of Lodz, Poland.
Cytokines, cellular & molecular therapy (ENGLAND) Mar **1999**, 5 (1) p15-23, ISSN 1368-4736 Journal Code: CUS
Languages: ENGLISH
Document type: Journal Article; Review; Review, Tutorial
Record type: Completed
Standard cytotoxic chemotherapy for neoplastic disease is fraught with systemic toxicity. The ratio of the toxic dose to the therapeutic dose is relatively low, which reflects the large number of cellular targets affected by the chemotherapeutic agent as well as its inability to distinguish between normal and malignant cells. The discovery of oncogenes and tumor suppressor genes involved in the process of transformation of normal cells into malignant cells has opened new areas of research in oncology, aimed at discovering drugs that could selectively inhibit their biological effects. This therapeutic modality, called an **antisense** strategy, has become a powerful tool for selectively reducing the expression of target genes in vitro, and there is increasing interest in the possibility of using the same technology in vivo for therapeutic purposes. In oncohematology, a number of trials have been initiated with **antisense** oligonucleotides directed against molecular targets, including the **bcl-2**, c-myc, bcr-abl, c-myb or p53 oncogenes and tumor suppressor genes. The experience gained from these studies will be applicable to the next generation of **antisense** compounds, which may include oligonucleotides with novel backbones or other structural modifications, as well as for expansion of the use of **antisense** oligonucleotides in combination approaches for the **treatment** of hematological malignancies.

4/3,AB/31 (Item 31 from file: 155)
DIALOG(R)File 155:MEDLINE(R)
(c) format only 2001 Dialog Corporation. All rts. reserv.

10247223 99381749 PMID: 10453946
Overexpression of **Bcl-2** protects human hepatoma cells from Fas-antibody-mediated apoptosis.
Takahashi M; Saito H; Okuyama T; Miyashita T; Kosuga M; Sumisa F; Yamada M; Ebinuma H; Ishii H
Department of Internal Medicine, School of Medicine, Keio University, Tokyo, Japan.
Journal of hepatology (DENMARK) Aug **1999**, 31 (2) p315-22, ISSN 0168-8278 Journal Code: IBS
Languages: ENGLISH
Document type: Journal Article
Record type: Completed
BACKGROUND/AIMS: Fas is a cell surface antigen, that triggers apoptosis upon specific ligand or antibody binding. The proto-oncogene **bcl-2** prevents apoptosis induced by various **treatments**. The aim of our study was to evaluate whether **Bcl-2** protects hepatoma cells from Fas-mediated apoptosis. METHODS: Two human cell lines, HCC-T and HepG2 were used. Expression of Fas antigen and **Bcl-2** was detected by flow cytometry and Western blotting. Cell viability and apoptotic change were examined after anti-Fas- and **antisense** oligodeoxynucleotide **treatments** . Apoptotic cells were detected by nick-end labelling and the TUNEL method. To test if **Bcl-2** expression can protect HepG2

cells from Fas-mediated apoptosis, the cells were transduced using retroviral vector, LZ[redacted] designed to coexpress E. coli beta-galactosidase and human **Bcl-2**. To further confirm the protective effect of **Bcl-2** expression against Fas-mediated apoptosis in HepG2, **Bcl-2** expressing plasmid vector was produced and a cell line stably expressing **Bcl-2** was cloned. RESULTS: Western blot analysis showed constitutive **Bcl-2** expression in HCC-T cells, but not in HepG2 cells. HCC-T was resistant to apoptosis after **treatment** with an agonist anti-Fas antibody (1 microg/ml for 3 days), whereas 33% of the HepG2 cells were killed by this **treatment**. Inhibition of **Bcl-2** expression by transfection of **antisense** oligodeoxynucleotides caused spontaneous apoptosis in HCC-T, but not in HepG2 cells, suggesting that **Bcl-2** is essential for survival of HCC-T cells, whereas other proteins may substitute for it in HepG2 cells. Following LZBC infection, 10% HepG2 cells were beta-galactosidase-positive by X-gal staining and **Bcl-2** -positive. In cells surviving after anti-Fas **treatment** , the proportion of beta-galactosidase-positive cells increased to 50% and the beta-galactosidase activity increased 6-fold, indicating that **Bcl-2** expression protected the cells from Fas-mediated apoptosis. In the cloned HepG2 cells stably expressing **Bcl-2** , the extent of Fas-mediated apoptosis was inversely related to the level of **Bcl-2** expression. CONCLUSION: **Bcl-2** confers protection to human hepatoma cells against Fas-mediated apoptosis, and is essential for survival of some, but not all, hepatoma cells.

4/3,AB/32 (Item 32 from file: 155)
DIALOG(R)File 155:MEDLINE(R)
(c) format only 2001 Dialog Corporation. All rts. reserv.

10244860 99391244 PMID: 10463603

Antisense Bcl-2 oligodeoxynucleotides inhibit progression to androgen-independence after castration in the Shionogi tumor model.

Miyake H; Tolcher A; Gleave ME
The Prostate Centre, Vancouver General Hospital, British Columbia, Canada.

Cancer research (UNITED STATES) Aug 15 **1999**, 59 (16) p4030-4, ISSN 0008-5472 Journal Code: CNF
Languages: ENGLISH
Document type: Journal Article
Record type: Completed

Progression to androgen-independence remains the main obstacle to improving survival for patients with advanced prostate cancer. Although **Bcl-2** expression in normal prostatic epithelial cells is low or absent, **Bcl-2** is highly up-regulated in prostate cancer cells after androgen withdrawal and during progression to androgen-independence. Here, we test the efficacy of **antisense Bcl-2** oligodeoxynucleotide (ODN) therapy administered adjuvantly after castration to delay time to androgen-independent recurrence in the androgen-dependent mouse Shionogi tumor model. **Treatment** of Shionogi tumor cells in vitro with **antisense Bcl-2** ODN inhibited **Bcl-2** expression in a dose-dependent and sequence-specific manner. Systemic administration of **antisense Bcl-2** ODN in mice bearing Shionogi tumors beginning 1 day postcastration resulted in a more rapid regression of tumors and a significant delay of emergence of androgen-independent recurrent tumors. Furthermore, despite significant reduction of **Bcl-2** expression in tumor tissues, **antisense Bcl-2** ODN had no effect on **Bcl-2** expression in normal mouse organs. These findings illustrate the potential utility of **antisense Bcl-2** therapy for prostate cancer in an adjuvant setting with androgen ablation.

4/3,AB/33 (Item 33 from file: 155)
DIALOG(R)File 155:MEDLINE(R)
(c) format only 2001 Dialog Corporation. All rts. reserv.

10233409 99369249 PMID: 10442640
Inhibition of Bcl-xL expression sensitizes normal human keratinocytes and epithelial cells to apoptotic stimuli.
Taylor JK; Zhang QQ; Monia BP; Marcusson EG; Dean NM
Department of Pharmacology, Isis Pharmaceuticals, Carlsbad, California 92008, USA.
Oncogene (ENGLAND) Aug 5 **1999**, 18 (31) p4495-504, ISSN 0950-9232 Journal Code: ONC
Languages: ENGLISH
Document type: Journal Article
Record type: Completed
The epidermis is continually exposed to harmful mutagens that have the potential to cause DNA damage. To protect the skin from accumulating mutated cells, keratinocytes have developed a highly regulated mechanism of eliminating damaged cells through apoptosis. Bcl-xL is a well-described cell survival protein that when overexpressed in skin can protect keratinocytes from UV radiation-induced apoptosis. To begin to unravel the complex mechanisms that keratinocytes use to survive, we wanted to characterize the role of endogenous Bcl-xL in protecting cells from death. In this study, we describe the development and characterization of an **antisense** inhibitor to Bcl-xL. We show that this inhibitor reduces Bcl-xL RNA and protein in a concentration-dependent, sequence-specific manner. Furthermore, **treatment** of keratinocytes and epithelial cells with this inhibitor sensitizes these cells to UV-B radiation and cisplatinum **treatment** -induced apoptosis. Thus, these results offer direct evidence that Bcl-xL is critical in the protection of skin and epithelial cells from apoptosis and provide a basis for the role of Bcl-xL in keratinocyte and epithelial cell survival.

4/3,AB/34 (Item 34 from file: 155)
DIALOG(R)File 155:MEDLINE(R)
(c) format only 2001 Dialog Corporation. All rts. reserv.

10204299 99253749 PMID: 10321832
Extracellular matrix inhibits apoptosis and enhances endothelial cell differentiation by a NfkappaB-dependent mechanism.
Wang W; Passaniti A
Laboratory of Biological Chemistry, National Institute on Aging, National Institutes of Health, Baltimore, Maryland 21224, USA.
Journal of cellular biochemistry (UNITED STATES) Jun 1 **1999**, 73 (3) p321-31, ISSN 0730-2312 Journal Code: HNF
Languages: ENGLISH
Document type: Journal Article
Record type: Completed
Hormonal and environmental factors that control the growth, differentiation, and regression of the vasculature are of fundamental importance in tumorigenesis and in the choice of therapeutic strategies. To test the hypothesis that estradiol (E2) and basement membrane proteins would affect the survival of vascular endothelial cells (EC), immortalized human umbilical vein endothelial cells (ECV304) were examined for their response to the chemotherapeutic drugs taxol and etoposide. ECV cell apoptosis was inhibited by E2 (taxol only) or attachment to extracellular matrix (ECM) (taxol or etoposide). E2 increased ECV growth, while ECM binding resulted in growth arrest and differentiation. Apoptosis was associated with decreased levels of **Bcl-2** and p21 proteins. E2 prevented down-regulation of p21 and **Bcl-2** induced by taxol but did not prevent the down-regulation of p21 induced by etoposide, consistent with the failure of E2 to inhibit etoposide-induced cell death. However, ECM prevented p21 and **Bcl-2** down-regulation induced by taxol or etoposide. Persistent activation of NFkappaB occurred after attachment of

ECV cells to ECM, suggesting a role in survival or differentiation. IkappaBalpha levels were not affected by taxol but were reduced by etoposide **treatment** , while IkappaBbeta levels did not change with drug **treatment** . E2 did not alter the levels of IkappaBalpha or IkappaBbeta. Interestingly, levels of IkappaBalpha and IkappaBbeta declined in etoposide-**treated** ECV cells on ECM concomitant with the elevation of NFkappaB, suggesting that in these cells degradation of IkappaB may be responsible for NFkappaB activation. In agreement with these data, anti-sense NFkappaB **treatment** of ECV cells inhibited differentiation on ECM, but did not affect cell survival. In conclusion, culture of ECV cells on ECM or **treatment** with E2 inhibited apoptosis. NFkappaB activation by ECM was necessary for cellular differentiation, rather than inhibition, of apoptosis.

4/3,AB/35 (Item 35 from file: 155)
DIALOG(R)File 155:MEDLINE(R)
(c) format only 2001 Dialog Corporation. All rts. reserv.

10197296 99326007 PMID: 10400420
Induction of apoptosis and differentiation by fludarabine in human leukemia cells (U937): interactions with the macrocyclic lactone bryostatin 1.

Vrana JA; Wang Z; Rao AS; Tang L; Chen JH; Kramer LB; Grant S
Department of Medicine, Medical College of Virginia, Virginia Commonwealth University, Richmond 23298, USA.

Leukemia (ENGLAND) Jul **1999**, 13 (7) p1046-55, ISSN 0887-6924
Journal Code: LEU
Contract/Grant No.: CA 63753, CA, NCI; CA77141, CA, NCI
Languages: ENGLISH
Document type: Journal Article
Record type: Completed

We have examined interactions between the purine nucleoside analog fludarabine (9-beta-arabinofuranosyl-2-fluoroadenine) and the macrocyclic lactone bryostatin 1 in the human monocytic leukemic cell line U937. Fludarabine exerted dose-dependent effects on U937 cell viability and growth which were associated with both induction of apoptosis, as well as cellular maturation. Incubation of cells with bryostatin 1 (10 nM; 24 h) after, but not before a 6-h exposure to 10 microM fludarabine resulted in a modest but significant increase in apoptosis, and was associated with greater than a 1 log reduction in clonogenicity. Subsequent exposure to bryostatin 1 also increased the percentage of fludarabine-**treated** cells displaying differentiation-related features (eg plastic adherence, CD11b positivity) compared to cells exposed to fludarabine alone. Bryostatin 1 did not increase the retention of the active fludarabine metabolite, F-ara-ATP, nor did it increase 3H-F-ara-A incorporation into DNA. Despite its capacity to trigger cellular maturation, fludarabine exposure (either with or without bryostatin 1) failed to induce the cyclin-dependent kinase inhibitors (CDKIs) p21WAF1/CIP1 and p27KIP1. Nevertheless, dysregulation of p21 (resulting from stable transfection of cells with a p21WAF1/CIP1 **antisense** construct) reduced fludarabine-mediated differentiation, while inducing a corresponding increase in apoptosis. Enforced expression of **Bcl-2** partially protected cells from fludarabine-related apoptosis, an effect that was overcome, in part, by subsequent exposure of cells to bryostatin 1. Interestingly, **Bcl-2** -overexpressing cells were as or in some cases, more susceptible to differentiation induction by fludarabine (+/- bryostatin 1) than their empty vector-containing counterparts. Collectively, these results indicate that the antiproliferative effects of fludarabine toward U937 leukemic cells involve both induction of apoptosis and cellular maturation, and that each of these processes may be enhanced by bryostatin 1.

4/3,AB/36 (Item 36 from file: 155)

DIALOG(R)File 155:MEDLINE(R)
(c) format only 2001 Dialog Corporation. All rts. reserv.

10185208 99238134 PMID: 10223618
CD40 stimulation inhibits cell growth and Fas-mediated apoptosis in a thyroid cancer cell line.
Fujieda S; Sugimoto C; Seki M; Sunaga H; Saito H
Department of Otorhinolaryngology, Fukui Medical University, Yoshida, Japan. sfujieda@fmsrsa.fukui-med.ac.jp
Oncology research (UNITED STATES) **1998**, 10 (9) p433-9, ISSN 0965-0407 Journal Code: BBN
Languages: ENGLISH
Document type: Journal Article
Record type: Completed
CD40 plays a critical role in the humoral immune response, especially in B-cell proliferation, differentiation, production of antibody, secretion of cytokines, and apoptosis. Here, we examined CD40 expression on six head and neck cancer cell lines by flow cytometry. Only the HTC/C3 cell line, which originated from a thyroid cancer, expressed CD40 on the surface of the cells. Coculture with anti-CD40 mAb inhibited colony formation of HTC/C3 cells. CD40 stimulation enhanced Fas expression on HTC/C3 cells. Although HTC/C3 cells are sensitive to Fas-mediated apoptosis, CD40 stimulation inhibited Fas-mediated apoptosis in HTC/C3 cells. CD40 stimulation enhanced **Bcl-2** expression, and **antisense** oligonucleotide against **bcl-2** canceled the inhibition of HTC/C3 cell growth caused by CD40 stimulation. Additionally, more anti-CD40 mAb-**treated** HTC/C3 cells were accumulated in G1 phase, and fewer in S phase, compared to nontreated cells. These results suggest that CD40 stimulation might be involved in the slow growth rate of CD40-bearing cancer cells, and they suggest a new biological approach to the **treatment** of cancers.

4/3,AB/37 (Item 37 from file: 155)
DIALOG(R)File 155:MEDLINE(R)
(c) format only 2001 Dialog Corporation. All rts. reserv.

10143922 99266307 PMID: 10333765
Antisense bcl-2 retrovirus vector increases the sensitivity of a human gastric adenocarcinoma cell line to photodynamic therapy.
Zhang WG; Ma LP; Wang SW; Zhang ZY; Cao GD
Institute of Biophysics, Academia Sinica, Beijing, China.
Photochemistry and photobiology (UNITED STATES) May **1999**, 69 (5) p582-6, ISSN 0031-8655 Journal Code: P69
Languages: ENGLISH
Document type: Journal Article
Record type: Completed
The **bcl-2** oncoprotein directly prolongs cellular survival by blocking apoptosis and its overexpression is associated with cellular resistance to killing by chemotherapeutic drugs and gamma-irradiation. Meanwhile, it has been shown that **bcl-2 antisense** oligonucleotide can induce apoptosis or increase toxicity of the **treatment** in tumors in vivo and in vitro. However, it is difficult to obtain stable transfection by this approach and there are no reports about the effect of an **antisense bcl-2** on the sensitivity to oxidative stress induced by photodynamic therapy (PDT). Here we investigated the effect of an **antisense bcl-2** RNA retrovirus vector transfer on the sensitivity of 2-butylamino-2-demethoxy-hypocrellin A (2-BA-2-DMHA) photosensitization in a human gastric adenocarcinoma MGC803 cell line. The results indicate that **antisense bcl-2**-infected MGC803 cells expressed exogenous **antisense bcl-2** mRNA measured by reverse transcription polymerase chain reaction and significantly reduced **bcl-2** protein determined by western blotting analysis. The decreased expression of **bcl-2** protein was accompanied by increased phototoxicity and

susceptibility to apoptosis induced by 2-BA-2-DMHA PDT. Our finding suggests that reduction of **bcl-2** protein in gastric cancers, and possibly also in a variety of other tumors, may be a novel and rational approach to improve photosensitivity and the **treatment** outcome.

4/3,AB/38 (Item 38 from file: 155)
DIALOG(R)File 155:MEDLINE(R)
(c) format only 2001 Dialog Corporation. All rts. reserv.

10070823 99164115 PMID: 10064615
Bcl-2 alters the balance between apoptosis and necrosis, but does not prevent cell death induced by oxidized low density lipoproteins.
Meilhac O; Escargueil-Blanc I; Thiers JC; Salvayre R; Negre-Salvayre A
INSERM U-466 and Department of Biochemistry and Molecular Biology, IFR-31, CHU Rangueil, Toulouse, France.
FASEB journal (UNITED STATES) Mar **1999**, 13 (3) p485-94, ISSN 0892-6638 Journal Code: FAS
Languages: ENGLISH
Document type: Journal Article
Record type: Completed
Oxidized low density lipoproteins (oxLDL) participate in atherosclerosis plaque formation, rupture, and subsequent thrombosis. Because oxLDL are toxic to cultured cells and **Bcl-2** protein prevents apoptosis, the present work aimed to study whether **Bcl-2** may counterbalance the toxicity of oxLDL. Two experimental model systems were used in which **Bcl-2** levels were modulated: 1) lymphocytes in which the (high) basal level of **Bcl-2** was reduced by **antisense** oligonucleotides; 2) HL60 and HL60/B (transduced by **Bcl-2**) expressing low and high **Bcl-2** levels, respectively. In cells expressing relatively high **Bcl-2** levels (lymphocytes and HL60/B), oxLDL induced mainly primary necrosis. In cells expressing low **Bcl-2** levels (**antisense-treated** lymphocytes, HL60 and ECV-304 endothelial cells), the rate of oxLDL-induced apoptosis was higher than that of primary necrosis. OxLDL evoked a sustained calcium rise, which is a common trigger to necrosis and apoptosis since both types of cell death were blocked by the calcium chelator EGTA. Conversely, a sustained calcium influx elicited by the calcium ionophore A23187 induced necrosis in cells expressing high **Bcl-2** levels and apoptosis in cells expressing low **Bcl-2** levels. This suggests that **Bcl-2** acts downstream from the calcium peak and inhibits only the apoptotic pathway, not the necrosis pathway, thus explaining the apparent shift from oxLDL-induced apoptosis toward necrosis when **Bcl-2** is overexpressed.

4/3,AB/39 (Item 39 from file: 155)
DIALOG(R)File 155:MEDLINE(R)
(c) format only 2001 Dialog Corporation. All rts. reserv.

10019779 99098950 PMID: 9880584
Contrasting role of presenilin-1 and presenilin-2 in neuronal differentiation in vitro.
Hong CS; Caromile L; Nomata Y; Mori H; Bredesen DE; Koo EH
Department of Neurosciences, University of California, San Diego, La Jolla, California 92093, USA.
Journal of neuroscience (UNITED STATES) Jan 15 **1999**, 19 (2) p637-43, ISSN 0270-6474 Journal Code: JDF
Contract/Grant No.: AG12282, AG, NIA; NS01812, NS, NINDS
Languages: ENGLISH
Document type: Journal Article
Record type: Completed
Presenilin-1 (PS1) and presenilin-2 (PS2), the major genes of familial Alzheimer's disease, are homologous to sel-12, a Caenorhabditis elegans gene involved in cell fate decision during development. Recently, wild-type

and mutant presenilins have been associated also with apoptotic cell death. By using stable transfection of **antisense** cDNAs, we studied the functions of PS1 and PS2 during neuronal differentiation in the NTera2 human teratocarcinoma (NT2) cell line. Expression of **antisense** PS1 resulted in a failure of the clones to differentiate into neurons after retinoic acid induction, whereas cells transfected with **antisense** PS2 differentiated normally. Concomitantly, **antisense** PS1 clones were associated with increased apoptosis both under basal conditions and during the early period of neuronal differentiation after retinoic acid **treatment**. Overexpression of **bcl-2** in **antisense** PS1 clones reduced cell death and resulted in a recovery of neuronal differentiation. These studies suggest that PS1 plays a role in differentiation and cell death and that PS1 and PS2 have differing physiological roles in this experimental paradigm.

4/3,AB/40 (Item 40 from file: 155)
DIALOG(R)File 155:MEDLINE(R)
(c) format only 2001 Dialog Corporation. All rts. reserv.

09954744 99054656 PMID: 9840921
Modulation of apoptosis by endogenous Bcl-xL expression in MKN-45 human gastric cancer cells.
Kondo S; Shinomura Y; Kanayama S; Higashimoto Y; Kiyohara T; Zushi S; Kitamura S; Ueyama H; Matsuzawa Y
Second Department of Internal Medicine, Osaka University Medical School, Suita, Japan.
Oncogene (ENGLAND) Nov 19 **1998**, 17 (20) p2585-91, ISSN 0950-9232 Journal Code: ONC
Languages: ENGLISH
Document type: Journal Article
Record type: Completed
This study was designed to clarify the role of endogenous Bcl-xL expression in modulating apoptosis of malignant cells. Administration of bcl-x-**antisense** oligonucleotides decreased Bcl-xL protein levels in the MKN-45 human gastric cancer cell line. The decrease in Bcl-xL protein content resulted in increased cell death induced by serum deprivation or Fas-antibody administration. Flow cytometric analysis revealed that the increased apoptotic cell death was more prominent in bcl-x-**antisense**-**treated** cells as compared to control cells, bcl-x-sense-**treated** cells, or bcl-x-nonsense-**treated** cells. To inhibit the effect of intrinsic Bcl-xL protein, we overexpressed Bak, which binds Bcl-xL and inhibits the anti-apoptotic effect of Bcl-xL, by transfection into MKN-45 cells. Bak-overexpressing cells showed increased apoptotic cell death induced by Fas-antibody when compared to parent cells and MKN-neo-transfected cells. Bak-overexpressing cells also showed greater sensitization to 5-fluorouracil and cisplatin than parent cells and MKN-neo-transfected cells. In conclusion, we demonstrated that administration of bcl-x-**antisense** oligonucleotides or overexpression of Bak protein induces sensitization to apoptosis in MKN-45 gastric cancer cells, suggesting that endogenous Bcl-xL expression in cancer cells is an important modulator of apoptosis.

4/3,AB/41 (Item 41 from file: 155)
DIALOG(R)File 155:MEDLINE(R)
(c) format only 2001 Dialog Corporation. All rts. reserv.

09948399 98451107 PMID: 9779827
Evidence of a direct role for **Bcl-2** in the regulation of articular chondrocyte apoptosis under the conditions of serum withdrawal and retinoic acid **treatment**.
Feng L; Precht P; Balakir R; Horton WE
Laboratory of Biological Chemistry, Gerontology Research Center, National Institute on Aging, NIH, Baltimore, Maryland 21224, USA.

Journal of cellular biochemistry (UNITED STATES) Nov 1 **1998**, 71 (2) p302-9, ISSN 073[redacted]312 Journal Code: HNF
Languages: ENGLISH
Document type: Journal Article
Record type: Completed

The regulation of chondrocyte apoptosis in articular cartilage may underlay age-associated changes in cartilage and the development of osteoarthritis. Here we demonstrate the importance of **Bcl-2** in regulating articular chondrocyte apoptosis in response to both serum withdrawal and retinoic acid **treatment** . Both stimuli induced apoptosis of primary human articular chondrocytes and a rat chondrocyte cell line as evidenced by the formation of DNA ladders. Apoptosis was accompanied by decreased expression of aggrecan, a chondrocyte specific matrix protein. The expression of **Bcl-2** was downregulated by both agents based on Northern and Western analysis, while the level of Bax expression remained unchanged compared to control cells. The importance of **Bcl-2** in regulating chondrocyte apoptosis was confirmed by creating cell lines overexpressing sense and **antisense Bcl-2** mRNA. Multiple cell lines expressing **antisense Bcl-2** displayed increased apoptosis even in the presence of 10% serum as compared to wild-type cells. In contrast, chondrocytes overexpressing **Bcl-2** were resistant to apoptosis induced by both serum withdrawal and retinoic acid **treatment**. Finally, the expression of **Bcl-2** did not block the decreased aggrecan expression in IRC cells **treated** with retinoic acid. We conclude that **Bcl-2** plays an important role in the maintenance of articular chondrocyte survival and that retinoic acid inhibits aggrecan expression independent of the apoptotic process.

4/3,AB/42 (Item 42 from file: 155)
DIALOG(R)File 155:MEDLINE(R)
(c) format only 2001 Dialog Corporation. All rts. reserv.

09918026 98435878 PMID: 9764849
Curcumin induces a p53-dependent apoptosis in human basal cell carcinoma cells.
Jee SH; Shen SC; Tseng CR; Chiu HC; Kuo ML
Department of Dermatology, College of Medicine, National Taiwan University, Taipei.
Journal of investigative dermatology (UNITED STATES) Oct **1998**, 111 (4) p656-61, ISSN 0022-202X Journal Code: IHZ
Languages: ENGLISH
Document type: Journal Article
Record type: Completed

Curcumin, a potent antioxidant and chemopreventive agent, has recently been found to be capable of inducing apoptosis in human hepatoma and leukemia cells by way of an elusive mechanism. Here, we demonstrate that curcumin also induces apoptosis in human basal cell carcinoma cells in a dose- and time-dependent manner, as evidenced by internucleosomal DNA fragmentation and morphologic change. In our study, consistent with the occurrence of DNA fragmentation, nuclear p53 protein initially increased at 12 h and peaked at 48 h after curcumin **treatment**. Prior **treatment** of cells with cycloheximide or actinomycin D abolished the p53 increase and apoptosis induced by curcumin, suggesting that either de novo p53 protein synthesis or some proteins synthesis for stabilization of p53 is required for apoptosis. In electrophoretic mobility gel-shift assays, nuclear extracts of cells **treated** with curcumin displayed distinct patterns of binding between p53 and its consensus binding site. Supportive of these findings, p53 downstream targets, including p21(CIP1/WAF1) and Gadd45, could be induced to localize on the nucleus by curcumin with similar p53 kinetics. Moreover, we immunoprecipitated extracts from basal cell carcinoma cells with different anti-p53 antibodies, which are known to be specific for wild-type or mutant p53 protein. The results reveal that basal cell carcinoma cells contain

exclusively wild-type p53; however, curcumin **treatment** did not interfere with cell cycling. Similarly, the apoptosis suppressor **Bcl-2** and promoter Bax were not changed with the curcumin **treatment**. Finally, **treatment** of cells with p53 **antisense** oligonucleotide could effectively prevent curcumin-induced intracellular p53 protein increase and apoptosis, but sense p53 oligonucleotide could not. Thus, our data suggest that the p53-associated signaling pathway is critically involved in curcumin-mediated apoptotic cell death. This evidence also suggests that curcumin may be a potent agent for skin cancer prevention or therapy.

4/3,AB/43 (Item 43 from file: 155)
DIALOG(R)File 155:MEDLINE(R)
(c) format only 2001 Dialog Corporation. All rts. reserv.

09907378 98414428 PMID: 9743471
Antisense inhibition of Bax mRNA increases survival of terminally differentiated HL60 cells.
Manfredini R; Capobianco ML; Trevisan F; Rauzi F; Barbieri D; Citro G; Tagliafico E; Ferrari S
Dipartimento di Scienze Biomediche, Sezione di Chimica Biologica, Universita di Modena, Italy.
Antisense & nucleic acid drug development (UNITED STATES) Aug **1998**, 8 (4) p341-50, ISSN 1087-2906 Journal Code: CJY
Languages: ENGLISH
Document type: Journal Article
Record type: Completed
Cell sensitivity to programmed cell death is primarily modulated by members of the **Bcl-2** family, as the balance of homodimer or heterodimer formation between proapoptotic and antiapoptotic members defines apoptosis susceptibility in the great majority of cellular contexts. It is, therefore, important to clarify if the Bax protein is limiting for activation of the genetic program of programmed cell death or can be complemented by different **Bcl-2** family members, such as Bak or Bad. To gain some insight into the role of Bax in the molecular mechanisms of apoptosis of myeloid cells, we inhibited this gene in all-trans-retinoic acid (ATRA)-**treated** HL60 cells using the methodology of **antisense** oligodeoxynucleotides (AS-ODN). Our results indicate that Bax inhibition has no effect on the proliferation and differentiation capacity of HL60 cells. Instead, the survival rate of terminally differentiated Bax-inactivated HL60 (Bax(-) HL60) cells is almost three times higher in respect to control cultures, indicating that in mature granulocytes Bax is not efficiently complemented by others members of the **Bcl-2** family proteins.

4/3,AB/44 (Item 44 from file: 155)
DIALOG(R)File 155:MEDLINE(R)
(c) format only 2001 Dialog Corporation. All rts. reserv.

09901089 99006633 PMID: 9792147
Synergistic cytotoxicity of **bcl-2** **antisense** oligodeoxynucleotides and etoposide, doxorubicin and cisplatin on small-cell lung cancer cell lines.
Zangemeister-Wittke U; Schenker T; Luedke GH; Stahel RA
Department of Internal Medicine, University Hospital Zurich, Switzerland.
British journal of cancer (SCOTLAND) Oct **1998**, 78 (8) p1035-42
, ISSN 0007-0920 Journal Code: AV4
Languages: ENGLISH
Document type: Journal Article
Record type: Completed
Expression of **Bcl-2** is life-sustaining for small-cell lung cancer cells and associated with drug resistance. In the present study, the interactions between the **bcl-2** **antisense**

oligodeoxynucleotide 2009 and the chemotherapeutic agents etoposide, doxorubicin and cisplatin were investigated on small-cell lung cancer cell lines to search for synergistic combinations. The cell lines NCI-H69, SW2 and NCI-H82 express high, intermediate-high and low basal levels of **Bcl-2**, respectively, which are inversely correlated with the sensitivities of the cell lines to **treatment** with oligodeoxynucleotide 2009 and the chemotherapeutic agents alone. Moreover, differences were found in the responsiveness of the cell lines to **treatment** with combinations of oligodeoxynucleotide 2009 and the chemotherapeutic agents. In the cell lines NCI-H69 and SW2, all combinations resulted in synergistic cytotoxicity. In NCI-H69 cells, maximum synergy with a combination index of 0.2 was achieved with the combination of oligodeoxynucleotide 2009 and etoposide. In SW2 cells, the combination of oligodeoxynucleotide 2009 and doxorubicin was the most effective (combination index = 0.5). In the cell line NCI-H82, which expresses a low basal level of **Bcl-2**, most of the combinations were slightly antagonistic. Our data suggest the use of oligodeoxynucleotide 2009 in combination with chemotherapy for the **treatment** of small-cell lung cancer that overexpresses **Bcl-2**.

4/3,AB/45 (Item 45 from file: 155)
DIALOG(R)File 155:MEDLINE(R)
(c) format only 2001 Dialog Corporation. All rts. reserv.

09839843 98384236 PMID: 9716601
Antisense to the epstein-barr virus (EBV)-encoded latent membrane protein 1 (LMP-1) suppresses LMP-1 and **bcl-2** expression and promotes apoptosis in EBV-immortalized B cells.
Kenney JL; Guinness ME; Curiel T; Lacy J
Department of Internal Medicine, Yale University School of Medicine, New Haven, CT, USA.
Blood (UNITED STATES) Sep 1 **1998**, 92 (5) p1721-7, ISSN 0006-4971 Journal Code: A8G
Contract/Grant No.: CA 67396, CA, NCI
Languages: ENGLISH
Document type: Journal Article
Record type: Completed
The Epstein-Barr virus (EBV)-encoded latent membrane protein (LMP-1) is required for viral transformation and functions to protect cells from apoptotic cell death, in part, by induction of antiapoptotic genes, including **Bcl-2** and A20. We have used **antisense** oligodeoxynucleotides targeted to LMP-1 as a strategy to suppress LMP-1 expression and thereby inhibit its functions. We have shown that levels of LMP-1 protein in EBV-positive lymphoblastoid cell lines can be reduced by in vitro **treatment** with unmodified oligodeoxynucleotides targeted to the first five codons of the LMP-1 open-reading frame. Furthermore, suppression of LMP-1 was associated with molecular and phenotypic effects that included downregulation of the LMP-1-inducible antiapoptotic genes, **Bcl-2** and Mcl-1, inhibition of proliferation, stimulation of apoptosis, and enhancement of sensitivity to the chemotherapeutic agent, etoposide. These effects were largely sequence-specific and observed in EBV-positive, but not EBV-negative cell lines. These studies suggest that lowering expression of LMP-1 in EBV-associated malignancy might have therapeutic effects and might synergize with other antitumor agents.
Copyright 1998 by The American Society of Hematology.

4/3,AB/46 (Item 46 from file: 155)
DIALOG(R)File 155:MEDLINE(R)
(c) format only 2001 Dialog Corporation. All rts. reserv.

09744798 98211344 PMID: 9551622
Antisense c-myc retroviral vector suppresses established human

prostate cancer.
Steiner MS; Anthony [redacted] Lu Y; Holt JT
Department of Urology, Vanderbilt University School of Medicine, Nashville, TN 37235, USA.
Human gene therapy (UNITED STATES) Mar 20 **1998**, 9 (5) p747-55, ISSN 1043-0342 Journal Code: A12
Languages: ENGLISH
Document type: Journal Article
Record type: Completed
Prostate cancer eventually becomes androgen resistant, resumes growth, and kills the patient. Characterization of genetic events that lead to androgen refractory prostatic neoplasia has revealed the frequent overexpression of c-myc and uncontrolled prostate cancer proliferation. A novel strategy to combat advanced prostate cancer utilized a replication incompetent retrovirus that contained the mouse mammary tumor virus (MMTV) promoter within the retroviral vector to allow transcription of **antisense** c-myc gene within target prostate tumor cells. The transduction of cultured DU145 cells by XM6:MMTV-**antisense** c-myc RNA retrovirus did not affect cell proliferation in culture, yet a single direct injection of MMTV-**antisense** c-myc viral media into established DU145 tumors in nude mice produced a 94.5% reduction in tumor size compared to tumors **treated** with control virus MTMV sense fos and untreated tumor by 70 days. Two animals in the **antisense** c-myc-**treated** group had complete regression of their tumors. Histopathological examination of the tumors revealed that MMTV-**antisense** c-myc-transduced DU145 tumors had increased tumor cell differentiation, decreased invasion, and a marked stromal response. The mechanism for the antitumor effect of MMTV-**antisense** c-myc retrovirus appears to be suppression of c-myc mRNA and protein, and decreased **bcl-2** protein. The in vivo transduction of prostate cancer cells with MMTV-**antisense** c-myc retroviruses reduced tumor growth by suppressing c-myc, resulting in the down-regulation of **bcl-2** protein. Consequently, the MMTV-**antisense** c-myc retrovirus may be useful for gene therapy against advanced, hormone-refractory prostate cancer.

4/3,AB/47 (Item 47 from file: 155)
DIALOG(R)File 155:MEDLINE(R)
(c) format only 2001 Dialog Corporation. All rts. reserv.

09734157 98240808 PMID: 9581676
Loss of butyrate-induced apoptosis in human hepatoma cell lines HCC-M and HCC-T having substantial **Bcl-2** expression.
Saito H; Ebinuma H; Takahashi M; Kaneko F; Wakabayashi K; Nakamura M; Ishii H
Department of Internal Medicine, School of Medicine, Keio University, Tokyo, Japan.
Hepatology (UNITED STATES) May **1998**, 27 (5) p1233-40, ISSN 0270-9139 Journal Code: GBZ
Languages: ENGLISH
Document type: Journal Article
Record type: Completed
We have demonstrated that sodium butyrate induces differentiation in human hepatoma cells; however, recent studies have shown that this agent causes apoptosis in some types of cancer cells. In this study, we examined whether sodium butyrate causes apoptosis in the human hepatoma cell lines, HCC-M and HCC-T. The growth of human hepatoma cells was dose-dependently reduced by sodium butyrate. Flow cytometric analysis showed cell-cycle arrest at the G1 phase in the sodium butyrate-**treated** cells. Apoptotic change was never found in **treated** cells at concentration levels of less than 5 mmol/L. Sodium butyrate decreased p53 expression and increased p21WAF-1 expression in HCC-T and HCC-M cells having the wild-type p53 gene. Western blot analysis showed that **Bcl-2** was expressed in the HCC-T and HCC-M cells, and its expression was increased after exposure to sodium butyrate. **Antisense** oligodeoxynucleotide against

bcl-2 easily caused apoptosis. These results indicate that sodium butyrate hardly induces apoptotic change in the human hepatoma cell lines, HCC-T and HCC-M, with the increase of **Bcl-2** expression. Cell-cycle arrest in the G1 phase caused by sodium butyrate was suggested to be induced by the increase in p21WAF-1 expression, but this change did not link with the p53 increase.

4/3,AB/48 (Item 48 from file: 155)
DIALOG(R)File 155:MEDLINE(R)
(c) format only 2001 Dialog Corporation. All rts. reserv.

09721662 98215921 PMID: 9547353
Cytotoxicity and apoptosis produced by cytochrome P450 2E1 in Hep G2 cells.
Chen Q; Cederbaum AI
Department of Biochemistry, Mount Sinai School of Medicine, New York 10029, USA.
Molecular pharmacology (UNITED STATES) Apr **1998**, 53 (4) p638-48, ISSN 0026-895X Journal Code: NGR
Contract/Grant No.: AA03312, AA, NIAAA; AA06610, AA, NIAAA
Languages: ENGLISH
Document type: Journal Article
Record type: Completed
Two Hep G2 subclones overexpressing CYP2E1 were established with the use of transfection and limited dilution screening techniques. The Hep G2-CI2E1-43 and -47 (E47) cells (transduced Hep G2 subclones that overexpress CYP2E1) grew at a slower rate than parental Hep G2 cells or control subclones that do not express CYP2E1, but remained fully viable. When GSH synthesis was inhibited by **treatment** with buthionine sulfoximine, GSH levels rapidly declined in E47 cells but not control cells, which is most likely a reflection of CYP2E1-catalyzed formation of reactive oxygen species. Under these conditions of GSH depletion, cytotoxicity and apoptosis were found only with the E47 cells. Low levels of lipid peroxidation were found in the E47 cells, which became more pronounced after GSH depletion. The antioxidants vitamin E, vitamin C, or trolox prevented the lipid peroxidation as well as the cytotoxicity and apoptosis, as did transfection with plasmid containing **antisense** CYP2E1 or overexpression of **Bcl-2**. Levels of ATP were lower in E47 cells because of damage to mitochondrial complex I. When GSH was depleted, oxygen uptake was markedly decreased with all substrates in the E47 extracts. Vitamin E completely prevented the decrease in oxygen uptake. Under conditions of CYP2E1 overexpression, two modes of CYP2E1-dependent toxicity can be observed in Hep G2 cells: a slower growth rate when cellular GSH levels are maintained and a loss of cellular viability when cellular GSH levels are depleted. Elevated lipid peroxidation plays an important role in the CYP2E1-dependent toxicity and apoptosis. This direct toxicity of overexpressed CYP2E1 may reflect the ability of this enzyme to generate reactive oxygen species even in the absence of added metabolic substrate.

4/3,AB/49 (Item 49 from file: 155)
DIALOG(R)File 155:MEDLINE(R)
(c) format only 2001 Dialog Corporation. All rts. reserv.

09694900 98184650 PMID: 9525737
Bcl-2 -independent Bcr-Abl-mediated resistance to apoptosis: protection is correlated with up regulation of Bcl-xL.
Amarante-Mendes GP; McGahon AJ; Nishioka WK; Afar DE; Witte ON; Green DR
Division of Cellular Immunology, La Jolla Institute for Allergy and Immunology, San Diego, California 92121, USA.
Oncogene (ENGLAND) Mar **1998**, 16 (11) p1383-90, ISSN 0950-9232 Journal Code: ONC
Languages: ENGLISH

Document type: Journal Article
Record type: Complete

Bcr - Abl is the molecule responsible for both the transformation phenotype and the resistance to chemotherapeutic drugs found in chronic myelogenous leukemia (CML) cells. Wild-type HL-60, a transformed pro-myelocytic cell line, is very susceptible to apoptosis-inducing agents. We show here that expression of Bcr - Abl in HL-60 cells rendered them extremely resistant to apoptosis induced by a wide variety of agents. The anti-apoptotic effect of Bcr - Abl was found to be independent of the phase of the cell cycle. **Treatment** with **antisense** oligonucleotides directed to bcr decreased the expression of the ectopic bcr - abl and restored susceptibility to apoptosis. Double mutations affecting the autophosphorylation site and the phosphotyrosine-binding motif (FLVRES) have been previously shown to impair the transforming activity of Bcr - Abl in fibroblasts and hematopoietic cells, however HL-60 cells expressing this double mutant molecule exhibited the same level of resistance to apoptosis as those expressing the wild-type Bcr - Abl. Interestingly, wild type and mutant Bcr - Abl induced in HL-60 cells a dramatic down regulation of **Bcl-2** and increased the levels of Bcl-xL. The level of Bax did not change in response to the presence of Bcr - Abl. **Antisense** oligonucleotides targeted to bcl-x downregulated the expression of Bcl-x, and increased the susceptibility of HL-60. Bcr - Abl cells to staurosporine. Importantly, HL-60 cells overexpressing Bcl-xL showed higher expression of Bcl-xL but lower resistance to apoptosis when compared to HL-60. Bcr - Abl cells. The results described here show that Bcr - Abl is a powerful mammalian anti-apoptotic molecule and can act independently of **Bcl-2** . Bcl-xL, however, seems to participate in part in Bcr - Abl-mediated resistance to apoptosis in HL-60 cells.

4/3,AB/50 (Item 50 from file: 155)
DIALOG(R)File 155:MEDLINE(R)
(c) format only 2001 Dialog Corporation. All rts. reserv.

09684125 98172985 PMID: 9514052

Growth inhibition of DU-145 prostate cancer cells by a **Bcl-2** **antisense** oligonucleotide is enhanced by N-(2-hydroxyphenyl)all-trans retinamide.

Campbell MJ; Dawson M; Koeffler HP
Division of Hematology/Oncology, Cedars-Sinai Medical Center/UCLA School of Medicine, Los Angeles, CA 90048, USA.

British journal of cancer (SCOTLAND) Mar **1998**, 77 (5) p739-44,
ISSN 0007-0920 Journal Code: AV4
Contract/Grant No.: CA42710, CA, NCI; CA43277, CA, NCI; CA70675-01, CA, NCI; +
Languages: ENGLISH
Document type: Journal Article
Record type: Completed

Hormonally insensitive prostate cancer is a relatively slow-growing, but usually fatal, disease with no long-term **treatment** options. Transformation of normal prostate cells to a malignant phenotype often involves corruption of the apoptotic machineries. **Bcl-2** protein is one of the key inhibitors of apoptosis and is often unregulated in advanced prostate cancer. The prostate cancer cell line DU-145 was used as a model of a hormonally insensitive, advanced prostate cancer. Cell growth in liquid culture was significantly inhibited by **antisense** **Bcl-2** oligonucleotides compared with control sense oligonucleotides; inhibition by these oligonucleotides was significantly enhanced on combination with the synthetic retinoid N-(2-hydroxyphenyl)all-trans-retinamide (2-HPR). Interestingly, growth inhibition occurred in the absence of apoptosis as measured using two assay techniques. We hypothesize that in these recalcitrant cells the apoptotic pathway is compromised at several levels, and **Bcl-2** may play another role in promoting cell growth. The use of **Bcl-2** **antisense** oligonucleotides plus 2-HPR may provide a novel approach to therapy of hormone-resistant prostate

cancer.

4/3,AB/51 (Item 51 from file: 155)
DIALOG(R)File 155:MEDLINE(R)
(c) format only 2001 Dialog Corporation. All rts. reserv.

09678215 98165184 PMID: 9506361
Differential induction of cell death in human glioma cell lines by sodium nitroprusside.
Blackburn RV; Galoforo SS; Berns CM; Motwani NM; Corry PM; Lee YJ
Department of Radiation Oncology, William Beaumont Hospital, Royal Oak, Michigan 48073, USA.
Cancer (UNITED STATES) Mar 15 **1998**, 82 (6) p1137-45, ISSN 0008-543X Journal Code: CLZ
Contract/Grant No.: CA 44550, CA, NCI; CA 48000, CA, NCI
Languages: ENGLISH
Document type: Journal Article
Record type: Completed
BACKGROUND: High grade gliomas represent very aggressive and lethal forms of human cancer, which often exhibit recurrence after surgical intervention and resistance to conventional chemotherapeutic and radiologic **treatment** . The clinically approved antihypertensive agent sodium nitroprusside (SNP) has been shown to induce cytotoxicity toward a number of carcinoma cell lines in vitro. METHODS: Three human glioma cell lines were examined for susceptibility to the cytotoxic effects of SNP. The role of the protein kinase C (PKC)alpha gene in mediating resistance to SNP-induced killing in U343 cells was investigated using **antisense** oligonucleotide inhibition. Stable transfection and overexpression of the PKCalpha gene in the SNP-susceptible cell line U251 was performed to further implicate PKCalpha as a mediating factor in SNP cytotoxicity. In addition, the presence of **bcl-2** protein in these cells was examined for possible correlation(s) with resistance to SNP. RESULTS: Exposure of U251 cells and LN-Z308 cells to 0.5 mM SNP resulted in significant cytotoxicity over a 72-hour period. U343 cells were resistant to SNP killing. U343 cells were shown to exhibit higher basal levels of PKCalpha and **bcl-2** than either U251 or LN-Z308 cells. **bcl-2** expression and resistance to SNP toxicity both were decreased by the introduction of PKCalpha **antisense** oligonucleotides into U343 cells. Conversely, enhanced PKC activity in PKCalpha-transfected U251 clones was associated with increased **bcl-2** expression and greater resistance to SNP-induced toxicity relative to control transfected cells. CONCLUSIONS: SNP can induce cytotoxicity in glioma cells. The susceptibility of these glioma cells to nitroprusside-induced killing appears to be correlated inversely with **bcl-2** and PKC activity. **bcl-2** levels in these cells can be altered through modulation of PKC signaling, specifically, by induction or inhibition of PKCalpha. These in vitro results provide an interesting basis for further study into the potential use of SNP for **treatment** of human gliomas in patients receiving combination therapy with conventional chemotherapeutic agents that exhibit PKC inhibitory activity.

4/3,AB/52 (Item 52 from file: 155)
DIALOG(R)File 155:MEDLINE(R)
(c) format only 2001 Dialog Corporation. All rts. reserv.

09673299 98119890 PMID: 9449720
The HIV-1 vpr protein acts as a negative regulator of apoptosis in a human lymphoblastoid T cell line: possible implications for the pathogenesis of AIDS.
Conti L; Rainaldi G; Matarrese P; Varano B; Rivabene R; Columba S; Sato A ; Belardelli F; Malorni W; Gessani S
Laboratory of Virology, Istituto Superiore di Sanita, Viale Regina Elena, 299-00161 Rome, Italy.

Journal of experimental medicine (UNITED STATES) Feb 2 **1998**, (3) p403-13, ISSN [redacted]-1007 Journal Code: I2V
Languages: ENGLISH
Document type: Journal Article
Record type: Completed

Although apoptosis is considered one of the major mechanisms of CD4(+) T cell depletion in HIV-infected patients, the virus-infected cells somehow appear to be protected from apoptosis, which generally occurs in bystander cells. Vpr is an auxiliary HIV-1 protein, which, unlike the other regulatory gene products, is present at high copy number in virus particles. We established stable transfectants of CD4+ T Jurkat cells constitutively expressing low levels of vpr. These clones exhibited cell cycle characteristics similar to those of control-transfected cells. **Treatment** of control clones with apoptotic stimuli (i.e., cycloheximide/tumor necrosis factor alpha (TNF-alpha), anti-Fas antibody, or serum starvation) resulted in a massive cell death by apoptosis. In contrast, all the vpr-expressing clones showed an impressive protection from apoptosis independently of the inducer. Notably, vpr **antisense** phosphorothioate oligodeoxynucleotides render vpr-expressing cells as susceptible to apoptosis induced by cycloheximide and TNF-alpha as the control clones. Moreover, the constitutive expression of HIV-1 vpr resulted in the upregulation of **bcl-2** , an oncogene endowed with antiapoptotic activities, and in the downmodulation of bax, a proapoptotic factor of the **bcl-2** family. Altogether, these results suggest that low levels of the endogenous vpr protein can interfere with the physiological turnover of T lymphocytes at early stages of virus infection, thus facilitating HIV persistence and, subsequently, viral spread. This might explain why apoptosis mostly occurs in bystander uninfected cells in AIDS patients.

4/3,AB/53 (Item 53 from file: 155)
DIALOG(R)File 155:MEDLINE(R)
(c) format only 2001 Dialog Corporation. All rts. reserv.

09671395 98143545 PMID: 9485014

1,25-Dihydroxyvitamin D3 protects human leukemic cells from tumor necrosis factor-induced apoptosis via inactivation of cytosolic phospholipase A2.

Wu YL; Jiang XR; Lillington DM; Allen PD; Newland AC; Kelsey SM
Department of Hematology, St. Bartholomew's and The Royal London School of Medicine and Dentistry, University of London, United Kingdom.

Cancer research (UNITED STATES) Feb 15 **1998**, 58 (4) p633-40, ISSN 0008-5472 Journal Code: CNF
Languages: ENGLISH
Document type: Journal Article
Record type: Completed

The mechanism by which tumor necrosis factor (TNF) induces death of cancer cells appears to involve the activation of cytosolic phospholipase A2 (cPLA2). U937 human leukemic cells **treated** with 1,25-dihydroxyvitamin D3 [1,25(OH)2D3; 10(-8) M] become resistant to TNF, an effect that is independent of cell cycle status and expression of TNF receptors or **BCL-2** . In this study, TNF produced a dose- and time-dependent enhancement of [3H]arachidonic acid release in U937 cells. The amount of [3H]arachidonic acid release was positively associated with TNF-induced apoptosis. Both immunofluorescence microscopy and Western blotting of cell subcompartments demonstrated translocation of cPLA2 from the cytosol to the cell membrane in response to TNF. In addition, TNF up-regulated expression of cPLA2 mRNA. An **antisense** oligonucleotide to cPLA2 and the cPLA2 inhibitor 4-bromophenacyl bromide significantly inhibited TNF-induced cytotoxicity. Prior incubation of cells with 1,25(OH)2D3 significantly inhibited (a) TNF-induced [3H]arachidonic acid release and apoptosis, (b) TNF-induced translocation of cPLA2 to the membrane, and (c) the up-regulation of cPLA2 mRNA with TNF. Furthermore, the inhibitory effect of 1,25(OH)2D3 was not reversed by inhibitors of

transcription or translation. The data suggest that activation of cPLA2 is involved in TNF-induced apoptosis of leukemic cells. 1[redacted](OH)2D3 directly inhibits cPLA2 translocation and mRNA up-regulation induced by TNF. Disruption of cPLA2 activation may represent a possible mechanism whereby leukemic cells can become resistant to TNF-mediated killing.

4/3,AB/54 (Item 54 from file: 155)
DIALOG(R)File 155:MEDLINE(R)
(c) format only 2001 Dialog Corporation. All rts. reserv.

09651495 98121097 PMID: 9461199
bcl-2 antisense therapy chemosensitizes human melanoma in SCID mice.
Jansen B; Schlagbauer-Wadl H; Brown BD; Bryan RN; van Elsas A; Muller M; Wolff K; Eichler HG; Pehamberger H
Department of Clinical Pharmacology, University of Vienna, Austria.
Nature medicine (UNITED STATES) Feb **1998**, 4 (2) p232-4, ISSN 1078-8956 Journal Code: CG5
Languages: ENGLISH
Document type: Journal Article
Record type: Completed
Malignant melanoma is a prime example of cancers that respond poorly to various **treatment** modalities including chemotherapy. A number of chemotherapeutic agents have been shown recently to act by inducing apoptosis, a type of cell death antagonized by the **bcl-2** gene. Human melanoma expresses **Bcl-2** in up to 90% of all cases. In the present study we demonstrate that **bcl-2 antisense** oligonucleotide **treatment** improves the chemosensitivity of human melanoma grown in severe combined immunodeficient (SCID) mice. Our findings suggest that reduction of **Bcl-2** in melanoma, and possibly also in a variety of other tumors, may be a novel and rational approach to improve chemosensitivity and **treatment** outcome.

4/3,AB/55 (Item 55 from file: 155)
DIALOG(R)File 155:MEDLINE(R)
(c) format only 2001 Dialog Corporation. All rts. reserv.

09635394 98082987 PMID: 9422524
Epstein-Barr virus LMP1 modulates the malignant potential of gastric carcinoma cells involving apoptosis.
Sheu LF; Chen A; Wei YH; Ho KC; Cheng JY; Meng CL; Lee WH
Department of Pathology, Tri-Service General Hospital, Taipei, Taiwan, Republic of China.
American journal of pathology (UNITED STATES) Jan **1998**, 152 (1) p63-74, ISSN 0002-9440 Journal Code: 3RS
Languages: ENGLISH
Document type: Journal Article
Record type: Completed
About 10% of gastric carcinomas including lymphoepithelioma-like carcinoma and adenocarcinoma are associated with Epstein-Barr virus (EBV) infection. In EBV-associated gastric carcinomas, the tumor cells express Epstein-Barr nuclear antigen 1 (EBNA-1) but not EBNA-2, -3A, -3B, or -3C, leader protein, or latent membrane proteins (LMPs) because of gene methylation. Only a few exceptional cases have LMP1 expression in tumor cells as demonstrated by immunohistochemical studies. To elucidate the biological effects of LMP1 and the significance of its restricted expression in EBV-associated gastric carcinomas, the LMP1 gene was transferred into EBV-negative gastric carcinoma cell lines (SCM1 and TMC1) and into EBV-negative nasopharyngeal carcinoma (NPC) cells (HONE-1) as a control. The biological effects of LMP1 in gastric carcinoma cells were monitored in vitro and in vivo. These results showed that the consequence of LMP1 expression is a growth enhancement in NPC cells, but it is a growth suppression in gastric carcinoma cells. The LMP1-expressing gastric

carcinoma cells had a reduced growth rate, colony-forming efficiency, mean colony size, and tumorigenicity and a lower malignant histological grade. The reduced growth rate, colony-forming efficiency, and mean colony size were partially reversible in vitro with **treatment** with LMP1 **antisense** oligonucleotide. In addition, enhanced apoptosis was found in the LMP1-expressing gastric carcinoma cells. This suggests that LMP1 may negatively modulate the malignant potential of gastric carcinoma cells via an enhancement of apoptosis. We concluded that the restriction of LMP1 expression in EBV-associated gastric carcinomas may lead to a growth advantage for tumor cells by avoiding LMP1 apoptotic effects and immunologically mediated elimination.

4/3,AB/56 (Item 56 from file: 155)
DIALOG(R)File 155:MEDLINE(R)
(c) format only 2001 Dialog Corporation. All rts. reserv.

09629924 98103643 PMID: 9443406
c-myc **antisense** oligodeoxynucleotides enhance the efficacy of cisplatin in melanoma chemotherapy in vitro and in nude mice.
Citro G; D'Agnano I; Leonetti C; Perini R; Bucci B; Zon G; Calabretta B; Zupi G
Laboratory of Experimental Chemotherapy, Regina Elena Cancer Institute, Centro Ricerca Sperimentale, Rome, Italy.
Cancer research (UNITED STATES) Jan 15 **1998**, 58 (2) p283-9,
ISSN 0008-5472 Journal Code: CNF
Languages: ENGLISH
Document type: Journal Article
Record type: Completed
This study was designed to assess the efficacy of a new antimelanoma therapeutic strategy that relies on the use of a c-myc **antisense** 15-mer phosphorothioate oligodeoxynucleotide ([S]ODN), in combination with cisplatin (cis-diamminedichloroplatinum; DDP), which is currently used in the clinical management of melanoma patients. Proliferation and colony formation of melanoma cells were both inhibited by the DDP/c-myc **antisense** [S]ODN combination to a greater extent than that observed with either agent alone. Inhibition was most effective when DDP was followed by c-myc **antisense** [S]ODNs. Cell cycle flow cytometric analysis of cells exposed to the two agents either alone or in combination demonstrated that (a) c-myc **antisense** [S]ODNs induced an accumulation of cells in S phase and apoptosis in a fraction of the cells, detectable at day 5 after the beginning of **treatment**; (b) DDP induced a block in G2-M phase detectable at day 1, which was partially recovered, and apoptosis similar in extent to that induced by c-myc **antisense** [S]ODNs; and (c) DDP and c-myc **antisense** [S]ODNs together induced arrest in G2-M phase, which was maximum at day 3, i.e., delayed as compared to the block induced by DDP. The combination induced a higher percentage of apoptosis, evident at day 3 from the start of **treatment**, that correlated with a marked reduction in **Bcl-2** expression. Mice bearing human melanoma xenografts and **treated** sequentially with DDP and c-myc **antisense** [S]ODNs showed a higher inhibition of tumor growth, reduction in the number of lung metastases, and increase in life span compared with those **treated** with either agent alone. Together, these data lend support to the development of anticancer therapies involving oncogene-targeted **antisense** ODNs and conventional antineoplastic drugs.

4/3,AB/57 (Item 57 from file: 155)
DIALOG(R)File 155:MEDLINE(R)
(c) format only 2001 Dialog Corporation. All rts. reserv.

09620439 98074644 PMID: 9413164
Development of a hammerhead **ribozyme** against **BCL-2**. II. **Ribozyme treatment** sensitizes hormone-resistant prostate cancer

cells to apoptotic agent[redacted]
Dorai T; Goluboff ET; [redacted]sson CA; Buttyan R
Department of Urology, College of Physicians and Surgeons, Columbia University, New York, NY 10032, USA.
Anticancer research (GREECE) Sep-Oct **1997**, 17 (5A) p3307-12,
ISSN 0250-7005 Journal Code: 59L
Languages: ENGLISH
Document type: Journal Article
Record type: Completed
BACKGROUND: Several lines of evidence strongly implicate a crucial role for the apoptosis suppressing **bcl-2** oncogene in the genesis of hormone-refractory human prostate cancer. By efficiently destroying the intracellular **bcl-2** mRNA, one might be able to make the prostate cancer cell responsive again to conventional apoptotic stimuli such as androgen withdrawal. To achieve this end, we have devised a catalytic **antisense** RNA strategy (**Ribozyme**) for **bcl-2** and evaluated its gene therapeutic potential. METHODS AND RESULTS: **Bcl-2** overexpressing LNCaP prostatic carcinoma cells (LNCaP/**bcl-2**) were transfected with the anti-**bcl-2** **ribozyme** RNA using a polyamine-based transfection reagent and the reduction in the intracellular **bcl-2** mRNA levels was followed by a ribonuclease protection assay. Using a cell viability assay, prior **ribozyme** transfection and subsequent application of apoptotic stimuli such as serum starvation or phorbol ester **treatment** caused a 30% increase in cell death by apoptosis than with these apoptotic stimuli alone. CONCLUSIONS: The results obtained strongly support the ability of a potential anti-**bcl-2** **ribozyme** therapy to synergize with other agents in inducing apoptosis of hormone-resistant human prostate cancer cells.

4/3,AB/58 (Item 58 from file: 155)
DIALOG(R)File 155:MEDLINE(R)
(c) format only 2001 Dialog Corporation. All rts. reserv.

09590455 97442404 PMID: 9295281
Protein kinase CbetaII activation by 1-beta-D-arabinofuranosylcytosine is antagonistic to stimulation of apoptosis and Bcl-2alpha down-regulation.
Whitman SP; Civoli F; Daniel LW
Department of Biochemistry, Bowman Gray School of Medicine, Wake Forest University, Winston-Salem, North Carolina 27157-1016, USA.
Journal of biological chemistry (UNITED STATES) Sep 19 **1997**, 272 (38) p23481-4, ISSN 0021-9258 Journal Code: HIV
Contract/Grant No.: CA43297, CA, NCI; CA48995, CA, NCI; CA67717, CA, NCI;
+
Languages: ENGLISH
Document type: Journal Article
Record type: Completed
1-beta-D-Arabinofuranosylcytosine (ara-C) stimulates the formation of both diglyceride and ceramide in the acute myelogenous leukemia cell line HL-60 (Strum, J. C., Small, G. W., Pauig, S. B., and Daniel, L. W. (1994) J. Biol. Chem 269, 15493-15497). ara-C also causes apoptosis in HL-60 cells which can be mimicked by exogenous ceramide. However, the signaling role for ara-C-induced diacylglycerol (DAG) is not defined. We found that **Bcl-2** levels were increased by **treatment** of HL-60 cells with exogenous DAG or 12-O-tetradecanoylphorbol-13-acetate (TPA). In contrast, exogenous ceramide **treatment** caused a decrease in cellular **Bcl-2** levels. Thus, ara-C stimulates the synthesis of two second messengers with opposing effects on **Bcl-2**. Since the effects of ara-C-induced DAG could be due to protein kinase C (PKC) activation, we determined the effects of ara-C on PKC isozymes. ara-C caused an increase in membrane-bound PKCbetaII (but not PKCalpha or PKCdelta). ara-C or TPA-induced translocation of PKCbetaII was inhibited by 1-O-octadecyl-2-O-methyl-rac-glycero-3-phosphocholine (ET-18-OCH3), and ara-C-induced apoptosis was stimulated by pretreatment of the cells with

ET-18-OCH3. ET-18-OCH3 also inhibited stimulation of **Bcl-2** by TPA and enhanced the decrease in **Bcl-2** observed in ara-C-**treated** cells. These data indicate that ara-C-induced apoptosis is limited by ara-C-stimulated PKCbetaII through effects on **Bcl-2**. To further determine the role of PKC, we used **antisense** oligonucleotides directed toward PKCbetaII. The **antisense**, but not the sense, oligonucleotide inhibited PKCbetaII activation and enhanced ara-C-induced apoptosis. These data demonstrate that the stimulation of apoptosis by ara-C is self-limiting and can be enhanced by inhibition of PKC.

4/3,AB/59 (Item 59 from file: 155)
DIALOG(R)File 155:MEDLINE(R)
(c) format only 2001 Dialog Corporation. All rts. reserv.

09572706 97434297 PMID: 9288183
Development of a hammerhead **ribozyme** against **bcl-2**. I. Preliminary evaluation of a potential gene therapeutic agent for hormone-refractory human prostate cancer.
Dorai T; Olsson CA; Katz AE; Buttyan R
Department of Urology, College of Physicians and Surgeons of Columbia University, New York, New York 10032, USA.
Prostate (UNITED STATES) Sep 1 **1997**, 32 (4) p246-58, ISSN 0270-4137 Journal Code: PB4
Languages: ENGLISH
Document type: Journal Article
Record type: Completed
BACKGROUND: The **bcl-2** oncoprotein suppresses apoptosis and, when overexpressed in prostate cancer cells, makes these cells resistant to a variety of therapeutic agents, including hormonal ablation. Therefore, **bcl-2** provides a strategic target for the development of gene knockout therapies to **treat** human prostate cancers. Towards this end, we have synthesized an anti-**bcl-2** gene therapeutic reagent based on **ribozyme** technology and have tested its effectiveness against **bcl-2** mRNA in vitro and in vivo. METHODS: A divalent hammerhead **ribozyme** was constructed by recombining two catalytic RNA domains into an **antisense** segment of the coding region for human **bcl-2** mRNA. A disabled **ribozyme** lacking catalytic activity was also constructed as a control reagent for our experiments. The **ribozymes** were tested for endonucleolytic activity against synthetic and natural **bcl-2** mRNAs. Simple transfection procedures were then utilized to introduce the **ribozymes** into cultured prostate cancer cells (LNCaP derivatives). We measured the effects of the **ribozymes** on endogenous expression of **bcl-2** mRNA and protein in these cells as well as their ability to induce apoptosis. RESULTS: The functional but not the disabled **ribozyme** was able to rapidly degrade **bcl-2** mRNA in vitro, without the requirement for any other cellular protein or factor. When directly transfected into LNCaP cell variants, it significantly reduced **bcl-2** mRNA and protein levels within 18 hr of **treatment**. This activity was sufficient to induce apoptosis in a low-**bcl-2** -expressing variant of LNCaP, but not in a high-**bcl-2**-expressing LNCaP line. For the high-**bcl-2**-expressing variant, however, it did restore the ability to genetically respond to a secondary apoptotic agent, phorbol ester, as evidenced by the renewed ability of phorbol ester to induce NGF1A mRNA in these cells. CONCLUSIONS: This study supports the potential utility of an anti-**bcl-2 ribozyme** reagent for reducing or eliminating **bcl-2** expression from hormone-refractory prostate cancer cells and for killing prostate cancer cells. As such, it is the first step toward an effective gene therapy against hormone-refractory human prostate cancers.

4/3,AB/60 (Item 60 from file: 155)

DIALOG(R)File 155:MEDLINE(R)
(c) format only 2001 Dialog Corporation. All rts. reserv.

09472925 98018463 PMID: 9380798
NSAID-induced apoptosis in Rous sarcoma virus-transformed chicken embryo fibroblasts is dependent on v-src and c-myc and is inhibited by **bcl-2**.
Lu X; Fairbairn DW; Bradshaw WS; O'Neill KL; Ewert DL; Simmons DL
Department of Zoology, Brigham Young University, Provo, UT 84602, USA.
Prostaglandins (UNITED STATES) Aug **1997**, 54 (2) p549-68, ISSN 0090-6980 Journal Code: Q76
Languages: ENGLISH
Document type: Journal Article
Record type: Completed
Mounting epidemiological and experimental evidence implicates non-steroidal antiinflammatory drugs as anti-tumorigenic agents. Our previous work showed that nonsteroidal antiinflammatory drug **treatment** of src-transformed chicken embryo fibroblasts caused apoptosis--a mechanism by which these drugs might exert their anti-tumorigenic effect. The present studies employ a sensitive technique for detecting single- and double-stranded DNA cleavage (the comet assay) to quantitate apoptosis. By this method pp60v-src, which antagonizes apoptosis in many cell systems, was found to induce apoptosis in 11-23% of serum-starved fibroblasts. However, **treatment** with diclofenac following pp60v-src activation produced a much stronger response beginning within 6 hours of **treatment** that resulted in 100% lethality. During cell death, cyclooxygenase-2 but not cyclooxygenase-1 mRNA was found to be uniformly increased by all apoptotic drugs tested. Examination of the expression of apoptosis-associated genes showed that c-rel and p53 (found in normal or v-src-transformed chicken embryo fibroblasts at moderate levels), and **bcl-2** (present at an extremely low level) were largely unchanged by **treatment** with eight different nonsteroidal antiinflammatory drugs. However, overexpression of human **bcl-2** inhibited diclofenac-mediated apoptosis by 90%, demonstrating directly that **bcl-2** expression can regulate nonsteroidal antiinflammatory drug induction of cell death. The proto-oncogene c-myc is known to cause apoptosis in chicken embryo fibroblasts when artificially overexpressed in cells deprived of trophic factors. We found that nonsteroidal antiinflammatory drug **treatment** following pp60v-src activation persistently induced myc protein and mRNA by more than 20-fold above that evoked by pp60v-src activation alone. Moreover, transfection of **antisense** c-myc oligonucleotides reduced drug-induced myc expression by 80% and caused a concomitant 50% reduction in cell death. These findings suggest that nonsteroidal antiinflammatory drug-induced apoptosis proceeds through a src/myc dependent pathway which is negatively regulated by **bcl-2**.

4/3,AB/61 (Item 61 from file: 155)
DIALOG(R)File 155:MEDLINE(R)
(c) format only 2001 Dialog Corporation. All rts. reserv.

09471017 98025896 PMID: 9376593
Cross-resistance of CD95- and drug-induced apoptosis as a consequence of deficient activation of caspases (ICE/Ced-3 proteases).
Los M; Herr I; Friesen C; Fulda S; Schulze-Osthoff K; Debatin KM
Hematology/Oncology, University Children's Hospital, Ulm, Germany.
Blood (UNITED STATES) Oct 15 **1997**, 90 (8) p3118-29, ISSN 0006-4971 Journal Code: A8G
Languages: ENGLISH
Document type: Journal Article
Record type: Completed
The cytotoxic effect of anticancer drugs has been shown to involve induction of apoptosis. We report here that tumor cells resistant to CD95 (APO-1/Fas) -mediated apoptosis were cross-resistant to apoptosis-induced

by anticancer drugs. Apoptosis induced in tumor cells by cytarabine, doxorubicin, and methotrexate required the activation of ICE/Ced-3 proteases (caspases), similarly to the CD95 system. After drug **treatment**, a strong increase of caspase activity was found that preceded cell death. Drug-induced activation of caspases was also found in ex vivo-derived T-cell leukemia cells. Resistance to cell death was conferred by a peptide caspase inhibitor and CrmA, a poxvirus-derived serpin. The peptide inhibitor was effective even if added several hours after drug **treatment**, indicating a direct involvement of caspases in the execution and not in the trigger phase of drug action. Drug-induced apoptosis was also strongly inhibited by **antisense** approaches targeting caspase-1 and -3, indicating that several members of this protease family were involved. CD95-resistant cell lines that failed to activate caspases upon CD95 triggering were cross-resistant to drug-mediated apoptosis. Our data strongly support the concept that sensitivity for drug-induced cell death depends on intact apoptosis pathways leading to activation of caspases. The identification of defects in caspase activation may provide molecular targets to overcome drug resistance in tumor cells.

4/3,AB/62 (Item 62 from file: 155)
DIALOG(R)File 155:MEDLINE(R)
(c) format only 2001 Dialog Corporation. All rts. reserv.

09456592 97151148 PMID: 8995684
Interaction of an adenovirus 14.7-kilodalton protein inhibitor of tumor necrosis factor alpha cytolysis with a new member of the GTPase superfamily of signal transducers.

Li Y; Kang J; Horwitz MS
Department of Microbiology and Immunology, Albert Einstein College of Medicine, Bronx, New York 10461, USA.

Journal of virology (UNITED STATES) Feb **1997**, 71 (2) p1576-82,
ISSN 0022-538X Journal Code: KCV
Contract/Grant No.: 5T32 CA09060, CA, NCI; P30-CA13330, CA, NCI
Languages: ENGLISH
Document type: Journal Article
Record type: Completed

The adenovirus (Ad) 14.7-kDa E3 protein (E3-14.7K), which can inhibit tumor necrosis factor alpha (TNF-alpha) cytolysis, was used to screen HeLa cell cDNA libraries for interacting proteins in the yeast two-hybrid system. A new member of the low-molecular-weight (LMW) GTP-binding protein family with Ras and ADP-ribosylation factor homology was discovered by this selection and has been named FIP-1 (14.7K-interacting protein). FIP-1 colocalized with Ad E3-14.7K in the cytoplasm especially near the nuclear membrane and in discrete foci on or near the plasma membrane. Its interaction with E3-14.7K was dependent on the FIP-1 GTP-binding domain. The stable expression of FIP-1 **antisense** message partially protected the cells from TNF-alpha cytolysis. FIP-1 was associated transiently with several unknown phosphorylated cellular proteins within 15 min after **treatment** with TNF-alpha. FIP-1 mRNA was expressed ubiquitously but at higher levels in human skeletal muscle, heart, and brain. In addition to homology to other LMW GTP-binding proteins, FIP-1 has regions of homology to two prokaryotic metalloproteases. However, there was no homology between FIP-1 and any of the recently isolated death proteins in the TNF-alpha or Fas/APO1 cytolytic pathway and no interaction with several members of the **Bcl-2** family of inhibitors of apoptosis. These data suggest that FIP-1, as a cellular target for Ad E3-14.7K, is either a new intermediate on a previously described pathway or part of a novel TNF-alpha-induced cell death pathway. FIP-1 has two consensus sequences for myristoylation which would be expected to facilitate membrane association and also has sequences for Ser/Thr as well as Tyr phosphorylation that could affect its function.

4/3,AB/63 (Item 63 from file: 155)
DIALOG(R)File 155:MEDLINE(R)
(c) format only 2001 Dialog Corporation. All rts. reserv.

09438108 98043628 PMID: 9373250
Thrombopoietin upregulates the promoter conformation of p53 in a proliferation-independent manner coincident with a decreased expression of Bax: potential mechanisms for survival enhancing effects.
Ritchie A; Gotoh A; Gaddy J; Braun SE; Broxmeyer HE
Departments of Microbiology/Immunology, Medicine (Hematology/Oncology), and the Walther Oncology Center, Indiana University School of Medicine, Indianapolis, IN, USA.
Blood (UNITED STATES) Dec 1 **1997**, 90 (11) p4394-402, ISSN 0006-4971 Journal Code: A8G
Contract/Grant No.: P01 HL 53586, HL, NHLBI; R01 HL 54037, HL, NHLBI; R01 HL 56416, HL, NHLBI; +
Languages: ENGLISH
Document type: Journal Article
Record type: Completed
Thrombopoietin (Tpo) has proliferative and maturational effects on immature and more committed cells, respectively. We previously reported a role for Tpo as a survival factor in the factor-dependent human cell line M07e by demonstrating that Tpo suppresses apoptosis in the absence of induced proliferation. Wild-type p53 is a tumor suppressor gene that can play a vital role in mediating growth factor withdrawal-induced apoptosis in factor-dependent hematopoietic cells. Wild-type p53 can switch from a suppressor conformation, with an antiproliferative, pro-apoptotic phenotype, to a promoter conformation that has a diminished ability to mediate cell cycle arrest and apoptosis. In an effort to elucidate the mechanisms through which Tpo suppresses apoptosis, we investigated the effects of Tpo **treatment** on p53-mediated apoptosis in M07e cells. Tpo upregulated the expression of the promoter conformation of p53 in M07e cells coincident with a downregulation of Bax and Mdm2 protein levels. Protein levels of **Bcl-2** and Bcl-xL did not significantly vary as a function of growth-factor stimulation. Conversely, the levels of suppressor conformation p53 were maximal when M07e was in a growth arrested state and decreased during factor stimulation. Furthermore, Tpo **treatment** induced an extranuclear buildup and greatly weakened the DNA binding capacity of p53. p53-specific **antisense** oligonucleotide **treatment** recapitulated the effects of Tpo **treatment** on the levels of Bax, Mdm-2, and **Bcl-2**. These results suggest that Tpo is suppressing growth factor withdrawal induced-apoptosis, at least in part, by downregulating the expression of pro-apoptotic Bax protein levels, through modulating the conformation of p53, which results in a functional inactivation of its pro-apoptotic abilities.

4/3,AB/64 (Item 64 from file: 155)
DIALOG(R)File 155:MEDLINE(R)
(c) format only 2001 Dialog Corporation. All rts. reserv.

09419583 98021579 PMID: 9378470
Antisense therapy for lymphomas.
Cotter FE
Molecular Haematology Unit, Institute of Child Health, London, U.K.
Hematological oncology (ENGLAND) Feb **1997**, 15 (1) p3-11, ISSN 0278-0232 Journal Code: GB2
Languages: ENGLISH
Document type: Journal Article; Review; Review, Tutorial
Record type: Completed
The potential ability of **antisense** oligonucleotides to downregulate the expression of oncogenes involved in lymphoma, with minimal toxicity can be achieved. The possibility of combining **antisense** therapy such as **BCL-2 antisense** with chemotherapy will probably provide an interesting means of overcoming tumour cell resistance to chemotherapy

in lymphoma and a range of other high **BCL-2** expressing malignancies. As additional **antisense** molecules targeting oncogenes involved in lymphomas become available, it will be possible to combine them with AO to enhance their efficacy, either targeting the same gene at two sites or more a combination of genes (for example, **BCL-2** and MYC in Burkitt's lymphoma). Of major importance are approaches to improve AO uptake into cells which is currently poor. Methods to improve **antisense** uptake into the cell are required and in addition a new generation of oligonucleotides free of the nonspecific thioate toxicities are required. AO are a dramatic new area of research and as such require much evaluation if they are to be applied maximally. Both in vitro and in vivo efficacy has been established. With care, novel therapies based on the biology of the malignant cell may be determined on a scientific basis and may help improve the **treatment** of patients with these diseases. Gene silencing by **antisense** oligonucleotides has a role to play as demonstrated in lymphomas.

4/3,AB/65 (Item 65 from file: 155)
DIALOG(R)File 155:MEDLINE(R)
(c) format only 2001 Dialog Corporation. All rts. reserv.

09398053 97267683 PMID: 9113013
BCL-2 antisense therapy in patients with non-Hodgkin lymphoma.
Webb A; Cunningham D; Cotter F; Clarke PA; di Stefano F; Ross P; Corbo M; Dziewanowska Z
Lymphoma Unit, Royal Marsden Hospital, Sutton, Surrey.
Lancet (ENGLAND) Apr 19 **1997**, 349 (9059) p1137-41, ISSN 0140-6736 Journal Code: L0S
Languages: ENGLISH
Document type: Clinical Trial; Clinical Trial, Phase I; Journal Article
Record type: Completed
BACKGROUND: Overexpression of **BCL-2** is common in non-Hodgkin lymphoma and leads to resistance to programmed cell death (apoptosis) and promotes tumorigenesis. **Antisense** oligonucleotides targeted at the open reading frame of the **BCL-2** mRNA cause a specific down-regulation of **BCL-2** expression which leads to increased apoptosis. Lymphoma grown in laboratory animals responds to **BCL-2 antisense** oligonucleotides with few toxic effects. We report the first study of **BCL-2 antisense** therapy in human beings. METHODS: A daily subcutaneous infusion of 18-base, fully phosporothioated **antisense** oligonucleotide was administered for 2 weeks to nine patients who had **BCL-2** -positive relapsed non-Hodgkin lymphoma. Toxicity was scored by the common toxicity criteria, and tumour response was assessed by computed tomography scan. Efficacy was also assessed by quantification of **BCL-2** expression; **BCL-2** protein levels were measured by flow cytometry in samples from patients. FINDINGS: During the course of the study, the daily dose of **BCL-2 antisense** was increased incrementally from 4.6 mg/m2 to 73.6 mg/m2. No **treatment**-related toxic effects occurred, apart from local inflammation at the infusion site. In two patients, computed tomography scans showed a reduction in tumour size (one minor, one complete response). In two patients, the number of circulating lymphoma cells decreased during **treatment** . In four patients, serum concentrations of lactate dehydrogenase fell, and in two of these patients symptoms improved. We were able to measure **BCL-2** levels by flow cytometry in the samples of five patients, two of whom had reduced levels of **BCL-2** protein. INTERPRETATION: In patients with relapsing non-Hodgkin lymphoma, **BCL-2 antisense** therapy led to an improvement in symptoms, objective biochemical and radiological evidence of tumour response, and down-regulation of the **BCL-2** protein in some patients. Our findings are encouraging and warrant further investigations of **BCL-2 antisense** therapy in cancer **treatment**.

4/3,AB/66 (Item 66 from file: 155)
DIALOG(R)File 155:MEDLINE(R)
(c) format only 2001 Dialog Corporation. All rts. reserv.

09397662 97250531 PMID: 9096387
Resistance to apoptosis in CTLL-2 cells constitutively expressing c-Myb is associated with induction of **BCL-2** expression and Myb-dependent regulation of **bcl-2** promoter activity.
Salomoni P; Perrotti D; Martinez R; Franceschi C; Calabretta B
Department of Microbiology and Immunology, Thomas Jefferson University, Philadelphia, PA 19107-6799, USA.
Proceedings of the National Academy of Sciences of the United States of America (UNITED STATES) Apr 1 **1997**, 94 (7) p3296-301, ISSN 0027-8424 Journal Code: PV3
Contract/Grant No.: R01 CA46782, CA, NCI
Languages: ENGLISH
Document type: Journal Article
Record type: Completed
c-Myb, the cellular homologue of the transforming gene of the avian myeloblastosis virus, is preferentially expressed in all hematopoietic lineages, including T and B lymphocyte lineages. In T lymphocytes, c-Myb expression appears to be required for cell cycle progression and proliferation. To further investigate the role of c-Myb in T cell proliferation and survival, interleukin (IL) 2-dependent CTLL-2 cells were transfected with a constitutively active c-myb or with a c-myb **antisense** construct able to down-regulate endogenous Myb levels, and the transfectants were assessed for proliferation and survival in low concentrations of IL-2 and for susceptibility to dexamethasone-induced apoptosis. Compared with control cells, CTLL-2 cells constitutively expressing c-Myb proliferate in low concentrations of IL-2 and are less susceptible to apoptosis induced by IL-2 deprivation or **treatment** with dexamethasone. In contrast, cells transfected with an **antisense** c-myb construct do not proliferate in low concentrations of IL-2 and undergo apoptosis upon IL-2 deprivation or dexamethasone **treatment** more rapidly than parental cells. Overexpression of c-Myb was accompanied by up-regulation of **BCL-2** expression. In transient transfection assays, the murine **bcl-2** promoter was efficiently transactivated by c-Myb, but such effect was observed also in cells transfected with a DNA binding-deficient c-myb construct. Moreover, in gel retardation assays, a 38-bp oligomer in the shortest **bcl-2** promoter segment regulated by c-Myb formed a specific complex with nuclear extracts from c-Myb-transfected CTLL-2 cells. Thus, these results strongly suggest that c-Myb, in addition to regulating T cell proliferation, protects T lymphocytes from apoptosis by induction of **BCL-2** expression, which involves a c-Myb-dependent mechanism of promoter regulation.

4/3,AB/67 (Item 67 from file: 155)
DIALOG(R)File 155:MEDLINE(R)
(c) format only 2001 Dialog Corporation. All rts. reserv.

09374324 97374435 PMID: 9230884
Induction of apoptosis in small-cell lung cancer cells by an **antisense** oligodeoxynucleotide targeting the **Bcl-2** coding sequence.
Ziegler A; Luedke GH; Fabbro D; Altmann KH; Stahel RA; Zangemeister-Wittke U
University Hospital Zurich, Department of Internal Medicine, Switzerland.
Journal of the National Cancer Institute (UNITED STATES) Jul 16 **1997**, 89 (14) p1027-36, ISSN 0027-8874 Journal Code: J9J
Comment in J Natl Cancer Inst. 1997 Jul 16;89(14) 988-90
Languages: ENGLISH

Document type: Journal Article
Record type: Complete
BACKGROUND: The emergence of resistance to chemotherapy remains a major problem in the **treatment** of patients with small-cell lung cancer. Elevated expression of **Bcl-2**, a protein that inhibits programmed cell death or apoptosis, has been associated with radiation and drug resistance and has been observed in the majority of small-cell lung cancer specimens and cell lines. PURPOSE: To test the hypothesis that **Bcl-2** expression levels are critical for inhibiting apoptosis in small-cell lung cancer cells, we used an **antisense** strategy to reduce **Bcl-2** expression in these cells in an attempt to restore the natural occurrence of apoptosis. METHODS: Thirteen **antisense** oligodeoxynucleotides (ODNs) targeting various regions of the **bcl-2** messenger RNA and a control scrambled-sequence ODN were tested to identify the most effective sequence(s) for reducing **Bcl-2** protein levels. Northern and western blot analyses were used to examine basal **bcl-2** messenger RNA and protein levels, respectively, in four human small-cell lung cancer cell lines (SW2, NCI-H69, NCI-H82, and NCI-N417). SW2 cells were **treated** with the **antisense** ODNs in the presence of cationic lipids (to facilitate uptake), and cytotoxic effects were measured by use of a cell viability assay. Flow cytometric analysis of DNA fragmentation and cell morphology was also performed. The cytotoxic effect of the most potent **antisense** ODN was also tested on the three other cell lines. RESULTS: The viability of SW2 cells was effectively reduced by ODNs that targeted the translation initiation and termination sites of the **bcl-2** messenger RNA, but ODN 2009 that targeted the coding region was the most cytotoxic. **Treatment** of SW2 cells with 0.15 microM ODN 2009 for 96 hours reduced their viability by 91% (95% confidence interval [CI] = 88%-94%) and caused a dose-dependent reduction in **Bcl-2** levels that became detectable 24 hours after **treatment** and persisted up to 96 hours; analysis of cellular morphology demonstrated that viability was reduced through apoptosis. Moreover, ODN 2009 at 0.15 microM was cytotoxic to NCI-H69, NCI-H82, and NCI-N417 cells, resulting in decreases in cell viability of 82% (95% CI = 78%-86%), 100%, and 100%, respectively, after 96 hours of **treatment**. The cytotoxic effects were inversely correlated with the basal **Bcl-2** levels in the cell lines (r = -9964). A control scrambled-sequence oligodeoxynucleotide had no statistically significant effect on the cell lines (P values ranging from .38 to .89). CONCLUSION: We have identified a novel **antisense** ODN sequence (ODN 2009) that effectively reduces the viability of small-cell lung cancer cells by reducing **Bcl-2** levels and facilitating apoptosis.

4/3,AB/68 (Item 68 from file: 155)
DIALOG(R)File 155:MEDLINE(R)
(c) format only 2001 Dialog Corporation. All rts. reserv.

09333438 97313416 PMID: 9169410
Cytotoxicity and apoptosis produced by arachidonic acid in Hep G2 cells overexpressing human cytochrome P4502E1.
Chen Q; Galleano M; Cederbaum AI
Department of Biochemistry, Mount Sinai School of Medicine, New York, New York 10029, USA.
Journal of biological chemistry (UNITED STATES) Jun 6 **1997**, 272 (23) p14532-41, ISSN 0021-9258 Journal Code: HIV
Contract/Grant No.: AA03312, AA, NIAAA; AA06610, AA, NIAAA
Languages: ENGLISH
Document type: Journal Article
Record type: Completed
The goal of the current study was to evaluate the effects of arachidonic acid, as a representative polyunsaturated fatty acid, on the viability of a Hep G2 cell line, which has been transduced to express human cytochrome P4502E1 (CYP2E1). Arachidonic acid produced a concentration- and time-dependent toxicity to Hep G2-MV2E1-9 cells, which express CYP2E1, but

little or no toxicity was found with control Hep G2-MV-5 cells, which were infected with retrovi[redacted] lacking human CYP2E1 cDNA. In contrast to arachidonic acid, oleic acid was not toxic to the Hep G2-MV2E1-9 cells. The cytotoxicity of arachidonic acid appeared to involve a lipid peroxidation type of mechanism since toxicity was enhanced after depletion of cellular glutathione; formation of malondialdehyde and 4-hydroxy-2-nonenal was markedly elevated in the cells expressing CYP2E1, and toxicity was prevented by antioxidants such as alpha-tocopherol phosphate, 6-hydroxy-2,5,7,8-tetramethylchroman-2-carboxylic acid (trolox), propylgall ate, ascorbate, and diphenylphenylenediamine, and the iron chelator desferrioxamine. Transfection of the Hep G2-MV2E1-9 cells with plasmid containing CYP2E1 in the sense orientation enhanced the arachidonic acid toxicity, whereas transfection with plasmid containing CYP2E1 in the **antisense** orientation decreased toxicity. The CYP2E1-dependent arachidonic acid toxicity appeared to involve apoptosis, as demonstrated by terminal deoxynucleotidyltransferase-mediated dUTP nick end labeling and DNA laddering experiments. Trolox, which prevented toxicity of arachidonic acid, also prevented the apoptosis. Transfection with a plasmid containing **bcl-2** resulted in complete protection against the CYP2E1-dependent arachidonic acid toxicity. It is proposed that elevated production of reactive oxygen intermediates by cells expressing CYP2E1 can cause lipid peroxidation, which subsequently promotes apoptosis and cell toxicity when the cells are enriched with polyunsaturated fatty acids such as arachidonic acid. The Hep G2-MV2E1-9 cells appear to be a valuable model to study interaction between CYP2E1, polyunsaturated fatty acids, reactive radicals, and the consequence of these interactions on cell viability and to reproduce several of the key features associated with ethanol hepatotoxicity in the intragastric infusion model of ethanol **treatment**.

4/3,AB/69 (Item 69 from file: 155)
DIALOG(R)File 155:MEDLINE(R)
(c) format only 2001 Dialog Corporation. All rts. reserv.

09321535 97246712 PMID: 9092950
Glucose deprivation-induced cytotoxicity in drug resistant human breast carcinoma MCF-7/ADR cells: role of c-myc and **bcl-2** in apoptotic cell death.
Lee YJ; Galoforo SS; Berns CM; Tong WP; Kim HR; Corry PM
Department of Radiation Oncology, William Beaumont Hospital, Royal Oak, Michigan 48073, USA.
Journal of cell science (ENGLAND) Mar **1997**, 110 (Pt 5) p681-6, ISSN 0021-9533 Journal Code: HNK
Contract/Grant No.: CA44550, CA, NCI; CA48000, CA, NCI
Languages: ENGLISH
Document type: Journal Article
Record type: Completed
We investigated the effect of glucose deprivation **treatment** on clonogenicity in multidrug-resistant human breast carcinoma MCF-7/ADR cells. Survival of MCF-7/ADR cells decreased exponentially up to 8 hours of incubation in the glucose-free medium. The surviving fraction of these cells for 8 hours of glucose-deprivation **treatment** was 1.5 x 10(-3). Photomicrographs and gel electrophoresis data suggest that glucose deprivation-induced cell death is associated with apoptosis. Data from western and northern blots showed an induction of c-myc gene expression during **treatment** with glucose-free medium in MCF-7/ADR cells. MCF-7/ADR cells transfected with c-myc **antisense** oligodeoxynucleotides became resistant to glucose deprivation-induced apoptosis. Overexpression of **bcl-2** gene protected MCF-7/ADR cells from this apoptotic cell death. Taken together, these results indicate that c-myc expression is a necessary component of glucose-free medium induced apoptosis and **bcl-2** prevents apoptotic death induced by c-myc.

4/3,AB/70 (Item 70 from file: 155)
DIALOG(R)File 155:MEDLINE(R)
(c) format only 2001 Dialog Corporation. All rts. reserv.

09295619 97275928 PMID: 9129689
Antisense oligonucleotides as therapeutics for malignant diseases.
Ho PT; Parkinson DR
Investigational Drug Branch, Cancer Therapy Evaluation Program, National Cancer Institute, Rockville, MD 20852, USA.
Seminars in oncology (UNITED STATES) Apr **1997**, 24 (2) p187-202
, ISSN 0093-7754 Journal Code: UN5
Languages: ENGLISH
Document type: Journal Article; Review; Review, Academic
Record type: Completed
The continued progress in our understanding of the biology of neoplasia and in the identification, cloning, and sequencing of genes critical to tumor cell function permits the exploitation of this information to develop specific agents that may directly modulate the function of these genes or their protein products. **Antisense** oligonucleotides are being investigated as a potential therapeutic modality that takes direct advantage of molecular sequencing. The **antisense** approach uses short oligonucleotides designed to hybridize to a target mRNA transcript through Watson-Crick base pairing. The formation of this oligonucleotide: RNA heteroduplex results in mRNA inactivation and consequent inhibition of synthesis of the protein product. A fundamental attraction of the **antisense** approach is that this method potentially may be applied to any gene product, in theory, for the **treatment** of malignant and non-malignant diseases. However, this simple and attractive model has proven to be much more complex in practice. A number of important challenges in the preclinical development of **antisense** oligonucleotides have been identified, including stability, sequence length, cellular uptake, target sequence selection, appropriate negative controls, oligonucleotide: protein interactions, and cost of manufacture. Although the biological activity of an oligonucleotide against its molecular target is theoretically sequence-dependent, the animal pharmacokinetics and toxicology of phosphorothioate analogues directed against vastly disparate gene products appear relatively non-sequence-specific. In oncology, a number of clinical trials have been initiated with **antisense** oligonucleotides directed against molecular targets including: p53; **bcl-2** ; raf kinase; protein kinase C-alpha; c-myb. The experience gained from these early clinical trials will be applicable to the next generation of **antisense** agents in development. These may include molecules with novel backbones or other structural modifications, chimeric oligonucleotides, or peptide nucleic acids. Continued progress in this arena will require that many of the preclinical challenges confronting **antisense** development are satisfactory resolved.

4/3,AB/71 (Item 71 from file: 155)
DIALOG(R)File 155:MEDLINE(R)
(c) format only 2001 Dialog Corporation. All rts. reserv.

09286158 97223337 PMID: 9123831
Expression of Epstein-Barr virus latent membrane protein 1 protects Jurkat T cells from apoptosis induced by serum deprivation.
Kawanishi M
Department of Microbiology, Graduate School of Medicine, Kyoto University, Japan.
Virology (UNITED STATES) Feb 17 **1997**, 228 (2) p244-50, ISSN 0042-6822 Journal Code: XEA
Languages: ENGLISH
Document type: Journal Article
Record type: Completed

It has been generally accepted that inhibition of apoptosis is important in the development of malignancy. To determine whether Epstein-Barr virus (EBV) latent membrane protein 1 (LMP1), the virus-coded transforming oncogene product, has an anti-apoptotic function in non-B-cells, Jurkat T cells were transfected with the LMP1-expression vector pSV2gptMTLM consisting of the human metallothionein promoter and were selected for mycophonolic acid resistance. LMP1-expressing clones of Jurkat cells showed resistance to apoptosis induced by serum deprivation. In LMP1-expressing clones, although the levels of **Bcl-2** and Bax were similar to those in the clones of vector transfectants or parental cells, c-Myc expression was significantly depressed. Down-regulation of c-Myc by LMP1 was confirmed by using LMP1-expressing clones **treated** with CdCl2. Addition of c-myc **antisense** oligonucleotides to Jurkat cells specifically inhibited apoptosis induced by serum deprivation at the concentrations which suppressed c-Myc expression. These results suggest that LMP1 expression and subsequent down-regulation of c-Myc protect Jurkat T cells from apoptosis induced by serum deprivation. The significance of the anti-apoptotic function of LMP1 in non-B, Jurkat T cells is discussed in relation to the pathogenesis of EBV malignancy.

4/3,AB/72 (Item 72 from file: 155)
DIALOG(R)File 155:MEDLINE(R)
(c) format only 2001 Dialog Corporation. All rts. reserv.

09283077 97258788 PMID: 9118846
Phenytoin-induced teratogenesis: a molecular basis for the observed developmental delay during neurulation.
Bennett GD; Lau F; Calvin JA; Finnell RH
Department of Veterinary Anatomy and Public Health, College of Veterinary Medicine, Texas A&M University, College Station 77843-4458, USA.
Epilepsia (UNITED STATES) Apr **1997**, 38 (4) p415-23, ISSN 0013-9580 Journal Code: EIX
Contract/Grant No.: DE11303, DE, NIDCR; ES07165, ES, NIEHS
Languages: ENGLISH
Document type: Journal Article
Record type: Completed
PURPOSE: We wished to determine whether chronic phenytoin (PHT) exposure could impair neural development and if any morphological alterations could be linked to changes in gene expression. METHODS: Pregnant SWV mice were chronically administered PHT 40 mg/kg/day from gestational day (GD) 0:12 (day:h) until they were killed at various timepoints throughout neural tube closure (NTC). At each timepoint, embryos from both **treated** and control dams were collected and scored for their progression through NTC. The neural tubes were then isolated and subjected to in situ transcription (IST) and **antisense** RNA amplification procedures. Using these techniques, we examined the expression of 10 genes: N-cadherin (Ncad), collagen type IV (col-IV), **bcl-2** , c-jun, PAX-3, collular retinol binding protein-2 (CRBP-2), retinoic acid receptor alpha (RAR alpha), transforming growth factor(beta2) (TGF(beta2)), wee-1, and EMX-2. RESULTS: Chronic PHT exposure not only caused a delay in NTC whereby exposed embryos lagged behind the controls at each collection timepoint, but also significantly altered the expression of specific genes at distinct times during NTC. Early in NTC, PHT induced a significant reduction in the expression of N-cad, col-IV, and c-jun in exposed embryos as compared with controls. In contrast, during the midstages of NTC, the only significant molecular alterations observed in the PHT-exposed embryos was the continued decreased expression of col-IV and an increase in CRBP-2 expression. Finally, in the latter stages of NTC, PHT caused a significant reduction in the expression of **bcl-2** , RAR alpha, TGF(beta2), EMX-2, and PAX-3. CONCLUSIONS: These results show that although the effects of PHT are morphologically subtle, causing a delay in the development of the neural tube, this delay is accompanied by alterations in critical genes at crucial times of neural development that may account for the observed neurological deficits often associated with PHT exposure.

4/3,AB/73 (Item 73 from file: 155)
DIALOG(R)File 155:MEDLINE(R)
(c) format only 2001 Dialog Corporation. All rts. reserv.

09274663 97213788 PMID: 9060477
Neuroprotective action of cycloheximide involves induction of **bcl-2** and antioxidant pathways.
Furukawa K; Estus S; Fu W; Mark RJ; Mattson MP
Sanders-Brown Research Center on Aging, University of Kentucky, Lexington 40536, USA.
Journal of cell biology (UNITED STATES) Mar 10 **1997**, 136 (5) p1137-49, ISSN 0021-9525 Journal Code: HMV
Contract/Grant No.: NS29001, NS, NINDS; NS30583, NS, NINDS
Languages: ENGLISH
Document type: Journal Article
Record type: Completed
The ability of the protein synthesis inhibitor cycloheximide (CHX) to prevent neuronal death in different paradigms has been interpreted to indicate that the cell death process requires synthesis of "killer" proteins. On the other hand, data indicate that neurotrophic factors protect neurons in the same death paradigms by inducing expression of neuroprotective gene products. We now provide evidence that in embryonic rat hippocampal cell cultures, CHX protects neurons against oxidative insults by a mechanism involving induction of neuroprotective gene products including the antiapoptotic gene **bcl-2** and antioxidant enzymes. Neuronal survival after exposure to glutamate, FeSO4, and amyloid beta-peptide was increased in cultures pretreated with CHX at concentrations of 50-500 nM; higher and lower concentrations were ineffective. Neuroprotective concentrations of CHX caused only a moderate (20-40%) reduction in overall protein synthesis, and induced an increase in c-fos, c-jun, and **bcl-2** mRNAs and protein levels as determined by reverse transcription-PCR analysis and immunocytochemistry, respectively. At neuroprotective CHX concentrations, levels of c-fos heteronuclear RNA increased in parallel with c-fos mRNA, indicating that CHX acts by inducing transcription. Neuroprotective concentrations of CHX suppressed accumulation of H2O2 induced by FeSO4, suggesting activation of antioxidant pathways. **Treatment** of cultures with an **antisense** oligodeoxynucleotide directed against **bcl-2** mRNA decreased **Bcl-2** protein levels and significantly reduced the neuroprotective action of CHX, suggesting that induction of **Bcl-2** expression was mechanistically involved in the neuroprotective actions of CHX. In addition, activity levels of the antioxidant enzymes Cu/Zn-superoxide dismutase, Mn-superoxide dismutase, and catalase were significantly increased in cultures exposed to neuroprotective levels of CHX. Our data suggest that low concentrations of CHX can promote neuron survival by inducing increased levels of gene products that function in antioxidant pathways, a neuroprotective mechanism similar to that used by neurotrophic factors.

4/3,AB/74 (Item 74 from file: 155)
DIALOG(R)File 155:MEDLINE(R)
(c) format only 2001 Dialog Corporation. All rts. reserv.

09186220 96292232 PMID: 8700536
A **bcl-2**/IgH **antisense** transcript deregulates **bcl-2** gene expression in human follicular lymphoma t(14;18) cell lines.
Capaccioli S; Quattrone A; Schiavone N; Calastretti A; Copreni E; Bevilacqua A; Canti G; Gong L; Morelli S; Nicolin A
Institute of General Pathology, University of Florence, Italy.
Oncogene (ENGLAND) Jul 4 **1996**, 13 (1) p105-15, ISSN 0950-9232
Journal Code: ONC
Languages: ENGLISH

Document type: Journal Article
Record type: Complete
The 14;18 chromosome translocation, characteristic of most human follicular B-cell lymphomas, juxtaposes the **bcl-2** gene with the IgH locus, creating a **bcl-2**/IgH hybrid gene. By mechanisms that are still under investigation, this event increases the cellular levels of the **bcl-2** mRNA and thereby induces an overproduction of the antiapoptotic **BCL-2** protein which is likely responsible for neoplastic transformation. In an effort to identify potential upregulators of **bcl-2** activity in t(14;18) cells, we found, by strand-specific RT-PCR, a **bcl-2 antisense** transcript that is present in the t(14;18) DOHH2 and SU-DHL-4 but not in the t(14;18)-negative Raji and Jurkat lymphoid cell lines, and thus appears to be dependent on the **bcl-2**/IgH fusion. This **antisense** transcript is a hybrid bcl-2/IgH RNA, that originates in the IgH locus, encompasses the t(14;18) fusion site and spans at least the complete 3' UTR region of the **bcl-2** mRNA. To achieve some insight into its biological function, we **treated** the t(14;18) DOHH2 cell line with oligonucleotides (ODNs) by specifically targeting the bcl-2/IgH **antisense** strand. These ODNs lowered **bcl-2** gene expression, inhibited neoplastic cell growth by inducing apoptosis. We would like to propose the hypothesis that the bcl-2/IgH **antisense** transcript may contribute, by an unknown mechanism, to upregulation of **bcl-2** gene expression in t(14;18) cells. The possibility has been considered that the hybrid **antisense** transcript mask AU-rich motifs present in the 3' UTR of the **bcl-2** mRNA characterized in other genes as mRNA destabilizing elements.

4/3,AB/75 (Item 75 from file: 155)
DIALOG(R)File 155:MEDLINE(R)
(c) format only 2001 Dialog Corporation. All rts. reserv.

09178556 97182982 PMID: 9031121
C-Myc and **Bcl-2** protein expression during the induction of apoptosis and differentiation in TNF alpha-**treated** HL-60 cells.
Kumakura S; Ishikura H; Tsumura H; Iwata Y; Endo J; Kobayashi S
Third Division of Internal Medicine, Shimane Medical University, Izumo, Japan.
Leukemia & lymphoma (SWITZERLAND) Oct **1996**, 23 (3-4) p383-94, ISSN 1042-8194 Journal Code: BNQ
Languages: ENGLISH
Document type: Journal Article
Record type: Completed
We examined c-Myc and **Bcl-2** protein expressions during the induction of apoptosis and differentiation in TNF alpha-**treated** HL-60 cells using a two-color flow cytometric method. We found that c-Myc protein was rapidly down-regulated in the apoptotic cells while **Bcl-2** protein was expressed at relatively high levels. Concomitantly with terminal differentiation **Bcl-2** protein was down-regulated in differentiating cells as well as c-Myc protein. We also showed that c-myc **antisense** oligonucleotides could induce apoptosis in HL-60 cells whereas **bcl-2 antisense** did not induce apoptosis during the early time of **treatment** . These results suggest that the down-regulation of c-Myc protein expression is a primary event to induce apoptosis and neither consistent expression of c-Myc protein nor rapid down-regulation of **Bcl-2** protein is necessary for the initial processing of apoptosis in HL-60 cells. Furthermore, concomitant down-regulation of c-Myc and **Bcl-2** is closely associated with terminal differentiation and apoptotic cell death of HL-60 cells **treated** with TNF alpha.

4/3,AB/76 (Item 76 from file: 155)
DIALOG(R)File 155:MEDLINE(R)

(c) format only 2001 Dialog Corporation. All rts. reserv.

09077814 96259035 PMID: 8984202
Antisense oligodeoxynucleotides to bax mRNA promote survival of rat sympathetic neurons in culture.
Gillardon F; Zimmermann M; Uhlmann E; Krajewski S; Reed JC; Klimaschewski L
II. Physiologisches Institut, Universitat Heidelberg, Germany.
Journal of neuroscience research (UNITED STATES) Mar 15 **1996**, 43 (6) p726-34, ISSN 0360-4012 Journal Code: KAC
Languages: ENGLISH
Document type: Journal Article
Record type: Completed

Previous in vitro studies have shown that the presence of high levels of Bax protein accelerated the rate of cell death following growth factor deprivation and that the ratio of cell death repressor **Bcl-2** to cell death effector Bax may determine the susceptibility to apoptosis. Both **Bcl-2** and Bax protein expression has been detected in sympathetic neurons in vivo, and overexpression of **bcl-2** in cultured sympathetic neurons prevented apoptosis after deprivation of nerve growth factor (NGF). In the present study, we investigated the expression of bax and **bcl-2** in primary cultures of sympathetic neurons from rat superior cervical ganglia. Furthermore, we tested the effects of a partially phosphorothioated bax **antisense** oligodeoxynucleotide (ODN) on the survival of sympathetic neurons in cultures supplied with suboptimal concentrations of NGF (0.5 ng/ml). A constitutive expression of bax mRNA at high levels was detected by reverse transcription and polymerase chain reaction which did not change significantly following NGF reduction or **treatment** with bax **antisense** ODN. A decrease in **Bcl-2** immunoreactivity was observed by immunocytochemistry in tyrosine hydroxylase-positive neurons when cultured under suboptimal NGF concentrations, whereas **Bcl-2** immunolabeled non-neuronal cells were not affected. Maximal number of neurons was obtained in control cultures containing 50 ng/ml of NGF. Few neurons survived in cultures grown in 0.5 ng/ml of NGF for 2 days (12.0 +/- 1.5% of controls, mean +/- SEM). Addition of two control ODNs at 1 microM had no effect on neuronal survival (10.1 +/- 1.2% and 11.0 +/- 1.3%, respectively), while the number of neurons was significantly increased in NGF-reduced cultures **treated** with a bax **antisense** ODNs (1 microM) (31.5 +/- 1.9%). Administration of fluorescein-labeled ODNs demonstrated intracellular uptake into cultured neurons. **Treatment** with bax **antisense** ODNs caused a significant reduction of Bax protein levels in SCG neurons by 46 +/- 2.6% as assessed by immuno-cytochemistry and digital image analysis. Taken together, our data demonstrate a constitutive expression of bax mRNA in sympathetic neurons suggesting that activation of bax expression may not be required for neuronal cell death after NGF withdrawal. After changing to suboptimal NGF concentrations, the cell-specific reduction in **Bcl-2** immunoreactivity preceded morphological signs of degeneration indicating that growth factor starvation may down-regulate neuronal **bcl-2** expression. **Treatment** with bax **antisense** ODNs indicated that suppression of Bax protein synthesis may promote neuronal survival in the threshold situation of insufficient trophic support.

4/3,AB/77 (Item 77 from file: 155)
DIALOG(R)File 155:MEDLINE(R)
(c) format only 2001 Dialog Corporation. All rts. reserv.

09050309 97070436 PMID: 8913362
Dopamine induces apoptotic cell death of a catecholaminergic cell line derived from the central nervous system.
Masserano JM; Gong L; Kulaga H; Baker I; Wyatt RJ
National Institute of Mental Health Neuroscience Center at Saint Elizabeths, Neuropsychiatry Branch, Washington, D.C. 20032, USA. masseraj@dirpc.nimh.nih.gov

Molecular pharmacology (UNITED STATES) Nov **1996**, 50 (5) p1309-15, ISSN 0026-8[redacted] Journal Code: NGR
Languages: ENGLISH
Document type: Journal Article
Record type: Completed

Dopamine produces a time- and dose-dependent increase in cell death in a clonal catecholaminergic cell line (CATH.a) derived from the central nervous system. Cell death also occurred after **treatment** with the catecholamines L-dihydroxyphenylalanine, norepinephrine, epinephrine, and isoproterenol, as well as the neurotoxic compound 6-hydroxydopamine. Cell death is not receptor mediated because selective noradrenergic and dopaminergic receptor agonists had no effect on CATH.a cell viability. Dopamine induces apoptotic cell death as indicated by DNA fragmentation measured by gel electrophoresis and by flow cytometric analysis. Apoptosis seems to be produced by dopamine autoxidation, because intracellular peroxides increase after dopamine **treatment** and cell death can be inhibited by catalase and N-acetylcysteine. N-acetylcysteine produced a dose-dependent decrease in dopamine-induced cell death; this correlated with a decrease in peroxide formation. In addition, **antisense** to the antioxidant protein **bcl-2** increases the sensitivity of CATH.a cells to dopamine-induced cell death. These findings indicate that the oxidative products of dopamine cause neurotoxicity through apoptosis.

4/3,AB/78 (Item 78 from file: 155)
DIALOG(R)File 155:MEDLINE(R)
(c) format only 2001 Dialog Corporation. All rts. reserv.

08974926 96319797 PMID: 8700802
Induction of apoptosis in prostatic tumor cell line DU145 by staurosporine, a potent inhibitor of protein kinases.
Zhang H; Hoang T; Saeed B; Ng SC
Department 4MG, Aging and Degenerative Diseases Research, Abbott Laboratories, Abbott Park, Illinois 60064, USA.
Prostate (UNITED STATES) Aug **1996**, 29 (2) p69-76, ISSN 0270-4137 Journal Code: PB4
Languages: ENGLISH
Document type: Journal Article
Record type: Completed

We are interested in studying the possibility of modulating prostatic cell growth by manipulating apoptosis. Here we show that 1 microM staurosporine (STS) induces a human androgen-independent prostatic tumor cell line, DU145, to undergo dramatic changes in morphology and results in programmed cell death. Several genes involved in apoptosis were analyzed for expression in STS-**treated** and untreated DU145 cells. It was observed that these genes were differentially regulated. The expression level of **bcl-2**, bcl-xL, Ich-1L remains unchanged in **treated** and untreated cells. On the other hand, DAD1 and interleukin-1 beta-converting enzyme (ICE) were downregulated while bcl-xs and Ich-1s were upregulated. By blocking **bcl-2** gene expression using **antisense** oligonucleotides, it was determined that the anti-**bcl-2** oligonucleotides have no effect on the proliferation of DU145 or STS-**treated** DU145 cells. These results demonstrate that programmed cell death can be induced in an androgen-independent prostatic cancer cell line and **BCL-2** was found not to play an important role in preventing STS-induced apoptosis in the DU145 cell line.

4/3,AB/79 (Item 79 from file: 155)
DIALOG(R)File 155:MEDLINE(R)
(c) format only 2001 Dialog Corporation. All rts. reserv.

08912036 96210674 PMID: 8633098
BCL2 regulates neural differentiation.
Zhang KZ; Westberg JA; Holtta E; Andersson LC

Department of Pathology, University of Helsinki, Haartman Institute, Finland.
Proceedings of the National Academy of Sciences of the United States of America (UNITED STATES) Apr 30 **1996**, 93 (9) p4504-8, ISSN 0027-8424 Journal Code: PV3
Languages: ENGLISH
Document type: Journal Article
Record type: Completed
A main function attributed to the BCL2 protein is its ability to confer resistance against apoptosis. In addition to the constitutively high expression of BCL2, caused by gene rearrangement in follicular lymphomas, elevated expression of the BCL2 gene has been found in differentiating hematopoietic, neural, and epithelial tissues. To address the question of whether the expression of BCL2 is a cause or consequence of cell differentiation, we used a human neural-crest-derived tumor cell line, Paju, that undergoes spontaneous neural differentiation in vitro. The Paju cell line displays moderate expression of BCL2, the level of which increases in parallel with further neural differentiation induced by **treatment** with phorbol 12-myristate 13-acetate. Transfection of normal human BCL2 cDNA in sense and **antisense** orientations had a dramatic impact on the differentiation of the Paju cells. Overexpression of BCL2 cDNA induced extensive neurite outgrowth, even in low serum concentrations, together with an increased expression of neuron-specific enolase. Paju cells expressing the anti-sense BCL2 cDNA construct, which reduced the endogenous levels of BCL2, did not undergo spontaneous neural differentiation. These cells acquired an epithelioid morphology and up-regulated the intermediate filament protein nestin, typically present in primitive neuroectodermal cells. The manipulated levels of BCL2 did not have appreciable impact on cell survival in normal culture. Our findings demonstrate that the BCL2 gene product participates in the regulation of neural differentiation.

4/3,AB/80 (Item 80 from file: 155)
DIALOG(R)File 155:MEDLINE(R)
(c) format only 2001 Dialog Corporation. All rts. reserv.

08875591 96224267 PMID: 8643560
Arachidonate lipoxygenases as essential regulators of cell survival and apoptosis.
Tang DG; Chen YQ; Honn KV
Department of Radiation Oncology, Wayne State University, Detroit, MI 48202, USA.
Proceedings of the National Academy of Sciences of the United States of America (UNITED STATES) May 28 **1996**, 93 (11) p5241-6, ISSN 0027-8424 Journal Code: PV3
Contract/Grant No.: CA29997, CA, NCI
Languages: ENGLISH
Document type: Journal Article
Record type: Completed
Arachidonic acid (AA) metabolites derived from both cyclooxygenase (COX) and lipoxygenase (LOX) pathways transduce a variety of signals related to cell growth. Here, we report that the AA LOX pathway also functions as a critical regulator of cell survival and apoptosis. Rat Walker 256 (W256) carcinosarcoma cells express 12-LOX and synthesize 12(S)- and 15(S)-hydroxyeicosatetraenoic acids as their major LOX metabolites. W256 cells transfected with 12-LOX-specific **antisense** oligonucleotide or **antisense** oligonucleotides directed to conserved regions of LOXs underwent time- and dose-dependent apoptosis. Likewise, **treatment** of W256 cells with various LOX but not COX inhibitors induced apoptotic cell death, which could be partially inhibited by exogenous 12(S)- or 15(S)-hydroxyeicosatetraenoic acids. The W256 cell apoptosis induced by **antisense** oligos and LOX inhibitors was followed by a rapid downregulation of **bcl-2** protein, a dramatic decrease in the **bcl-2**/bax ratio, and could be suppressed by **bcl-2**

overexpression. In contrast, p53, which is wild type in W256 cells, did not undergo alterations during apoptosis induction. The results suggest that the LOX pathway plays an important physiological role in regulating apoptosis.

4/3,AB/81 (Item 81 from file: 155)
DIALOG(R)File 155:MEDLINE(R)
(c) format only 2001 Dialog Corporation. All rts. reserv.

08872375 96220013 PMID: 8639245
Oligonucleotides induce apoptosis restricted to the t(14;18) DHL-4 cell line.
Morelli S; Alama A; Quattrone A; Gong L; Copreni E; Canti G; Nicolin A
Department of Pharmacology, University of Milan, Italy.
Anti-cancer drug design (ENGLAND) Jan **1996**, 11 (1) p1-14,
ISSN 0266-9536 Journal Code: AC5
Languages: ENGLISH
Document type: Journal Article
Record type: Completed
Most human follicular B-cell lymphomas are associated with t(14;18) chromosome translocation that joins the **bcl-2** gene with the IgH locus. This hybrid gene causes upregulation of **BCL-2** protein expression, endowing cells with survival advantage. Although early **BCL-2** overexpression is definitely responsible for immortalization/transformation, its exact role in the overt transformation as well as in the maintenance of the tumor phenotype is not known. The capacity of oligodeoxynucleotides (ODN) to modulate gene expression specifically has been exploited to downregulate the overexpression of **BCL-2** protein in the SU-DHL-4 human follicular B-cell lymphoma line by the use of sense ODN or **antisense** ODN or **antisense** ODN designed to encompass the unique nucleotide sequence in the fusion region of the hybrid transcript. The specific downregulation of the **bcl-2** transcript and of the relevant **BCL-2** protein in the **treated** cells activated programed cell death and inhibited growing cells. The antitumor activity was restricted to the DHL-4 cell line carrying the specific nucleotide sequence at the **bcl-2**/IgH joining region. Thus, DHL-4 lymphoma cells derived from the acute phase of human follicular B-cell lymphoma, although endowed with additional activated oncogenes, were growth inhibited by **bcl-2** downregulation with additional activated oncogenes, were growth inhibited by **bcl-2** downregulation in a genetically restricted fashion. The biological activity was exerted exclusively by ODNs synthesized in the sense orientation. The sense ODNs have been proposed to anneal the hybrid **bcl-2**/IgH **antisense** RNA as identified in this study.

4/3,AB/82 (Item 82 from file: 155)
DIALOG(R)File 155:MEDLINE(R)
(c) format only 2001 Dialog Corporation. All rts. reserv.

08867014 96185047 PMID: 8605968
Stromal cells regulate **bcl-2** and bax expression in pro-B cells.
Gibson LF; Piktel D; Narayanan R; Nunez G; Landreth KS
Department of Pediatrics, West Virginia University Health Sciences Center, Morgantown, USA.
Experimental hematology (UNITED STATES) Apr **1996**, 24 (5) p628-37, ISSN 0301-472X Journal Code: EPR
Contract/Grant No.: AI23950, AI, NIAID
Languages: ENGLISH
Document type: Journal Article
Record type: Completed
B lymphocyte production in the bone marrow depends on a cascade of regulatory cells and cytokines unique to the hematopoietic

microenvironment. Fibroblastic stromal cells appear to be particularly important in regulating [redacted] earliest events in this line[redacted]; however, it is still not clear whether the same or different sets of signals regulate maintenance of cell viability, proliferation, and differentiation of B lineage cells. In this study, we addressed the role of bone marrow stromal cells in survival and expansion of normal murine pro-B cells. Stromal cells were required for long-term proliferation of pro-B cell clone C1.92, and, in the presence of stromal cell line S10, pro-B cells expressed the proto-oncogene **bcl-2** . Removal of C1.92 cells from Stromal cell-derived signaling in support of pro-B cell viability. Due to its previously described role in regulating cell survival, we investigated whether stromal cells regulate **bcl-2** expression in pro-B cells. When removed from stromal cell cultures, pro-B cells rapidly lost **bcl -2** mRNA expression coincident with initiation of apoptosis. However, interruption of **bcl-2** expression with **antisense** oligonucleotides in the presence of stroma and interleukin-7 (IL-7) did not result in immediate cell death. Oligonucleotide-**treated** cells arrested in G(1) phase of the cell cycle 24 hours before the initiation of apoptosis. In contrast, removal of pro-B cells from stromal cell support resulted in rapid increase in BAX expression, correlating directly with initiation of apoptosis. These results suggest that **bcl-2** may, in part, regulate cell survival by interrupting the cascade of intracellular events that regulate cell cycle progression in lymphopoietic cells. Initiation of apoptosis in these cells appears to be more closely correlated with intracellular levels of BAX expression.

4/3,AB/83 (Item 83 from file: 155)
DIALOG(R)File 155:MEDLINE(R)
(c) format only 2001 Dialog Corporation. All rts. reserv.

08804677 96051283 PMID: 8528964
Antisense oligodeoxyribonucleotide down-regulation of **bcl-2** gene expression inhibits growth of the low-grade non-Hodgkin's lymphoma cell line WSU-FSCCL.
Smith MR; Abubakr Y; Mohammad R; Xie T; Hamdan M; al-Katib A
Department of Medical Oncology, Fox Chase Cancer Center, Philadelphia, Pennsylvania 19111, USA.
Cancer gene therapy (UNITED STATES) Sep **1995**, 2 (3) p207-12,
ISSN 0929-1903 Journal Code: CE3
Languages: ENGLISH
Document type: Journal Article
Record type: Completed
The **BCL-2** gene product is involved in preventing apoptosis. The t(14,18) chromosomal translocation, which results in a fusion messenger RNA containing the entire coding region of **BCL-2** and a portion of the immunoglobulin heavy chain gene, is commonly found in follicular lymphoma and appears to play a role in lymphomagenesis by inhibiting cell death. We tested the hypothesis that downregulation of **BCL-2** would decrease accumulation of follicular lymphoma cells by **treating** the t(14,18)-carrying follicular lymphoma cell line WSU-FSCCL in vitro with **antisense** oligodeoxyribonucleotides (ODNs) directed against **BCL -2** . We found dose-dependent, sequence-specific inhibition of cell accumulation by an **antisense** unmodified ODN directed at codons 2 to 7, which downregulated **BCL-2** protein levels. This effect was near maximal at an ODN concentration of 40 micrograms/mL (6.9 mumol/L), with minimal toxicity by control sense, reverse, and mutated **antisense** ODN at the same concentration. The pre-B leukemia cell line REH showed no sequence-specific growth inhibition by the **antisense** ODN at these concentrations, and **BCL-2** protein levels were not altered. These data suggest that WSU-FSCCL may be useful in a murine model to optimize **antisense** ODN for potential therapeutic utility.

4/3,AB/84 (Item 84 from file: 155)

DIALOG(R)File 155:MEDLINE(R)
(c) format only 2001 Dialog Corporation. All rts. reserv.

08793795 95129488 PMID: 7828537
Inhibitors of oxidative stress mimic the ability of follicle-stimulating hormone to suppress apoptosis in cultured rat ovarian follicles.
Tilly JL; Tilly KI
Department of Population Dynamics, Johns Hopkins University, Baltimore, Maryland 21205-2179.
Endocrinology (UNITED STATES) Jan **1995**, 136 (1) p242-52,
ISSN 0013-7227 Journal Code: EGZ
Contract/Grant No.: 5 P30 HD-06268-21, HD, NICHD
Languages: ENGLISH
Document type: Journal Article
Record type: Completed
We have reported that members of the **bcl-2** gene family are expressed and gonadotropin regulated in ovarian granulosa cells during follicular maturation and atresia. Because **Bcl-2**, a protein that prevents apoptosis in several cell types, is reported to function as an antioxidant or free radical scavenger, the present studies were designed to investigate if oxidative stress plays a role in granulosa cell apoptosis during follicular atresia in the immature rat ovary. In the first series of experiments, the role of oxidative stress in the induction of granulosa cell apoptosis was directly tested using a defined in vitro follicle culture system. Healthy antral follicles obtained from equine CG (eCG)-primed immature (27 day old) rats were incubated in serum-free medium for 24 h in the absence or presence of FSH (100 ng/ml; a control for inhibiting apoptosis), superoxide dismutase (SOD; 10-1000 U/ml), ascorbic acid (0.01-1 mM; a free radical scavenger), N-acetyl-L-cysteine (25-100 mM; a free radical scavenger and stimulator of endogenous glutathione peroxidase activity), or catalase (10-1000 U/ml). Granulosa cells within follicles incubated in medium alone exhibited extensive apoptosis after 24 h of incubation, and this onset of apoptosis was blocked by **treatment** with FSH (29 +/- 4% of controls; $P < 0.001$, $n = 3$). Moreover, apoptosis in follicles was also inhibited by **treatment** with SOD (44 +/- 4% of controls at 1000 U/ml; $P < 0.01$, $n = 3$), ascorbic acid (55 +/- 9% of controls at 1 mM; $P < 0.05$, $n = 3$), N-acetyl-L-cysteine (24 +/- 7% of controls at 100 mM; $P < 0.001$, $n = 3$), or catalase (35 +/- 6% of controls at 1000 U/ml; $P < 0.001$, $n = 3$). In the second series of experiments, complementary DNAs corresponding to secreted (SEC-SOD), copper/zinc-containing (Cu/Zn-SOD), and manganese-containing (Mn-SOD) forms of rat SOD, rat seleno-cysteine glutathione peroxidase (GSHPx), and rat catalase were isolated and used to synthesize **antisense** RNA probes for Northern and slot blot analysis of changes in SOD, GSHPx, and catalase gene expression during follicular maturation. In vivo priming of 25-day-old female rats for 2 days with 10 IU eCG, which promoted antral follicular growth and survival, increased levels of messenger RNA encoding SEC-SOD (216 +/- 9% of saline-**treated** controls, $P < 0.05$, $n = 3$) and Mn-SOD (222 +/- 14% of saline-**treated** controls, $P < 0.05$, $n = 3$) vs. saline-**treated** controls.(ABSTRACT TRUNCATED AT 400 WORDS)

4/3,AB/85 (Item 85 from file: 155)
DIALOG(R)File 155:MEDLINE(R)
(c) format only 2001 Dialog Corporation. All rts. reserv.

08739341 95368651 PMID: 7641210
Estrogen promotes chemotherapeutic drug resistance by a mechanism involving **Bcl-2** proto-oncogene expression in human breast cancer cells.
Teixeira C; Reed JC; Pratt MA
Department of Pharmacology, University of Ottawa, Ontario, Canada.
Cancer research (UNITED STATES) Sep 1 **1995**, 55 (17) p3902-7,
ISSN 0008-5472 Journal Code: CNF
Languages: ENGLISH

Document type: Journal Article
Record type: Complete

Recent studies have shown that the **Bcl-2** protein suppresses programmed cell death or apoptosis induced by a variety of stimuli including chemotherapeutic drugs. Because estrogen promotes the survival of estrogen-dependent breast cancer cells in vivo, we investigated whether estrogen might regulate levels of **Bcl-2** gene expression in an estrogen-responsive human breast cancer cell line. Estrogen receptor-positive MCF-7 human breast cancer cells cultured in the presence of estrogen express the 8.5-kb **Bcl-2** mRNA transcript. Depletion of estrogen from the medium results in loss of expression of the mRNA, whereas reexposure to estrogen markedly induces the **Bcl-2** transcript. The changes in **Bcl-2** mRNA are paralleled by changes in **Bcl-2** protein levels. Estrogen-induced increases in **Bcl-2** are significantly inhibited by inclusion of the pure antiestrogen ICI 164,384 in the medium. The Bax protein that heterodimerizes with **Bcl-2** and promotes cell death is expressed in MCF-7 cells grown in the presence of estrogen and is unaffected by culture in estrogen-free medium. Estrogen depletion doubles the sensitivity of MCF-7 cells to the cytotoxic effects of Adriamycin compared with cells cultured in medium supplemented with estrogen, consistent with a decrease in the **Bcl-2** levels. MCF-7 cells **treated** simultaneously with estrogen and ICI 164,384 exhibit markedly lower resistance to Adriamycin compared with cells **treated** with estrogen alone. In the absence of estrogen, MCF-7 cells transfected with **Bcl-2** expression plasmids display a marked increase in resistance to Adriamycin. In the presence of estrogen, MCF-7 cells expressing **Bcl-2** **antisense** transcripts are rendered twice as sensitive to acute Adriamycin cytotoxicity as a control clone. We conclude that estrogen can promote resistance of estrogen receptor bearing human breast cancer cells to chemotherapeutic drugs through a mechanism that involves regulation of the **Bcl-2** proto-oncogene.

4/3,AB/86 (Item 86 from file: 155)
DIALOG(R)File 155:MEDLINE(R)
(c) format only 2001 Dialog Corporation. All rts. reserv.

08737698 95315480 PMID: 7795171
The role of **Bcl-2** protein and autocrine growth factors in a human follicular lymphoma-derived B cell line.
Blagosklonny MV; Neckers LM
Clinical Pharmacology Branch, National Cancer Institute, National Institute of Health, Bethesda, MD 20892, USA.
European cytokine network (FRANCE) Jan-Feb **1995**, 6 (1) p21-7,
ISSN 1148-5493 Journal Code: A56
Languages: ENGLISH
Document type: Journal Article
Record type: Completed

We have shown that the ability of the human follicular lymphoma-derived cell line SU-DHL-6 to proliferate and survive in vitro depends on both **Bcl-2** expression and multiple autocrine growth factors. **Treatment** with **Bcl-2** **antisense** (AS **Bcl-2**) decreased **Bcl-2** protein levels. However, a cytotoxic effect was seen only at very restricted cell densities. Below such densities cells underwent spontaneous death without any **treatment**, while above these cell densities no cytotoxic effect of AS **Bcl-2** could be seen. The conditioned medium of SU-DHL cells supported the survival and growth of these cells cultivated at low cell densities and partially reversed the cytotoxicity associated with **Bcl-2** depletion. RT/PCR analysis revealed autocrine expression of IL-1 beta, IL-2, IL-5 and TNF-beta in SU-DHL cells. Neutralizing antibodies against these cytokines inhibited SU-DHL proliferation. Thus, development of autocrine GF secretion may be the second step in the pathogenesis of follicular lymphomas.

4/3,AB/87 (Item 87 from file: 155)
DIALOG(R)File 155:MEDLINE(R)
(c) format only 2001 Dialog Corporation. All rts. reserv.

08734868 95204963 PMID: 7897244
A simple assay for examining the effect of transiently expressed genes on programmed cell death.
Memon SA; Petrak D; Moreno MB; Zacharchuk CM
Laboratory of Immune Cell Biology, National Cancer Institute, National Institutes of Health, Bethesda, MD 20892-1152.
Journal of immunological methods (NETHERLANDS) Mar 13 **1995**, 180 (1) p15-24, ISSN 0022-1759 Journal Code: IFE
Languages: ENGLISH
Document type: Journal Article
Record type: Completed
Programmed cell death (PCD) has been observed in a wide variety of cell types in response to physiologic signals or types of stress. How these stimuli trigger PCD, and whether there is a common PCD signal transduction pathway, is not clear. As more genes are described that may participate in or regulate PCD, an assay system in which gene products can easily be introduced and/or modulated would be of great value. To avoid the generation and screening of multiple individual stable cell transfectants, a simple transient transfection death assay has been developed. 2B4.11, a murine T cell hybridoma, was transfected by electroporation with a constitutively active beta-galactosidase reporter gene and the cells were incubated in culture medium or with a PCD-inducing stimulus. The amount of beta-galactosidase activity remaining in the intact cells at the end of the culture period represented only viable transfected cells. **Bcl-2** was chosen to examine whether this system would be useful to study the effect of transiently transfected genes since it blocks PCD in a number of experimental systems. Consistent with data obtained using stable transfectants, transient expression of **Bcl-2** in 2B4.11 completely protected cells from glucocorticoid- and cytotoxic agent-induced PCD. This protection from death was confirmed at the individual cell level by the transient co-expression of a class I Ld surface antigen and flow cytometric analysis. Some of the advantages of the transient transfection death assay described here are; (1) the simple and sensitive beta-galactosidase assay, (2) the rapidity of the assay, (3) the ability to perform conventional viability assays to monitor **treatment**-induced cytotoxicity, (4) multiple gene products can be tested alone, and in combination, (5) **antisense** or dominant negative approaches can be used, and (6) the adaptability of this assay system to other cell types, transfection techniques, or reporter and expression vectors. The transient transfection death assay should make it easier to identify and order important steps in the PCD signal transduction pathways.

4/3,AB/88 (Item 88 from file: 155)
DIALOG(R)File 155:MEDLINE(R)
(c) format only 2001 Dialog Corporation. All rts. reserv.

08734128 95153658 PMID: 7850782
Androgens induce resistance to **bcl-2**-mediated apoptosis in LNCaP prostate cancer cells.
Berchem GJ; Bosseler M; Sugars LY; Voeller HJ; Zeitlin S; Gelmann EP
Department of Medicine, Lombardi Cancer Center, Georgetown University School of Medicine, Washington, DC 20007.
Cancer research (UNITED STATES) Feb 15 **1995**, 55 (4) p735-8, ISSN 0008-5472 Journal Code: CNF
Contract/Grant No.: CA57176, CA, NCI
Languages: ENGLISH
Document type: Journal Article
Record type: Completed

We describe an in vitro model for prostate cancer **treatment** that suggests a potential benefit for combined androgen ablation and cytotoxic chemotherapy. Androgen **treatment** of the LNCaP hormone-dependent human prostate cancer cell line induces increased expression of the **BCL-2** protein. Increased levels of this protein are known to mediate inhibition of apoptosis. LNCaP cells, however, did not undergo apoptosis in response to androgen withdrawal. Etoposide exerts its cytotoxicity on LNCaP and other cells by inducing apoptosis. In vitro etoposide cytotoxicity was diminished 83% in the presence of either 10(-8) M dihydrotestosterone or 10(-9) M R1881 in LNCaP cells. The interaction between androgen and etoposide was mediated through the **BCL-2** protein, since **bcl-2** **antisense** oligonucleotides blocked the protective effect of androgens on etoposide cytotoxicity.

4/3,AB/89 (Item 89 from file: 155)
DIALOG(R)File 155:MEDLINE(R)
(c) format only 2001 Dialog Corporation. All rts. reserv.

08529236 95293939 PMID: 7775402
Expression of differentiation-related phenotypes and apoptosis are independently regulated during myeloid cell differentiation.
Terui Y; Furukawa Y; Sakoe K; Ohta M; Saito M
Division of Hemopoiesis, Institute of Hematology, Jichi Medical School, Tochigi.
Journal of biochemistry (JAPAN) Jan **1995**, 117 (1) p77-84,
ISSN 0021-924X Journal Code: HIF
Languages: ENGLISH
Document type: Journal Article
Record type: Completed
When human promyelocytic leukemia cell line HL-60 was **treated** with various differentiation-inducers, apoptosis always occurred after the full appearance of differentiation-related phenotypes. However, the two phenomena could be dissociated when HL-60 cells were **treated** with PDBu. When HL-60 cells were cultured with PDBu for more than 36 h, apoptosis was induced following differentiation. Apoptosis was not, however, observed when PDBu was removed within 24 h, even though induction of differentiation-related phenotypes, such as NBT-reducing ability and surface marker expression, was the same as that in the control. Northern blot analysis revealed that **bcl-2** mRNA was rapidly down-regulated within 6 h of the **treatment** with PDBu. The amount of **bcl-2** mRNA recovered to that of undifferentiated HL-60 cells when PDBu was washed out within 24 h. In contrast, the recovery of **bcl-2** was incomplete when the cells were **treated** with PDBu for more than 36 h, suggesting that **bcl-2** is also a critical regulator of the cell fate during myeloid differentiation. This hypothesis was confirmed by experiments using **antisense** oligonucleotides, i.e., blocking the recovery of **bcl-2** mRNA by **antisense** oligonucleotides could result in the induction of apoptosis in HL-60 cells from which PDBu was removed within 24 h. Moreover, overexpression of **BCL-2** in HL-60 cells could block apoptosis during differentiation without any significant effect on differentiation itself. These results strongly suggest that apoptosis is not a simple consequence of differentiation-induction, and that apoptosis and differentiation are regulated independently in myeloid cells.

4/3,AB/90 (Item 90 from file: 155)
DIALOG(R)File 155:MEDLINE(R)
(c) format only 2001 Dialog Corporation. All rts. reserv.

08436831 94358414 PMID: 7521364
Involvement of LFA-1/intracellular adhesion molecule-1-dependent cell adhesion in CD40-mediated inhibition of human B lymphoma cell death induced by surface IgM crosslinking.

Sumimoto S; Heike T; [redacted]nazashi S; Shintaku N; Jung EY; [redacted]Hata D; Katamura K ; Mayumi M
Department of Pediatrics, Faculty of Medicine, Kyoto University, Japan.
Journal of immunology (UNITED STATES) Sep 15 **1994**, 153 (6) p2488-96, ISSN 0022-1767 Journal Code: IFB
Languages: ENGLISH
Document type: Journal Article
Record type: Completed

B cells have been shown to receive negative signals for their growth through crosslinking of surface IgM (sIgM), and it has been demonstrated that anti-IgM Abs induce B cell death. Proliferation of B cells in response to Ag stimulation in vivo may thus require additional signals that inhibit the sIgM-transduced negative signals. Signaling through CD40 has been proposed as a candidate for such costimulatory signals. To investigate the role of CD40-transduced signals in sIgM-mediated B cell death, we used a human B cell line (DND-39) that expresses sIgM, sIgD, and CD40. Crosslinking of sIgM, but not sIgD, by Abs induced DND-39 cell death. The dying cells showed the morphology of apoptosis and DNA fragmentation. Anti-CD40 Abs induced homotypic adhesion of DND-39 cells and rescued them from anti-IgM Ab-induced cell death. Anti-CD40 Abs inhibited anti-IgM Ab-induced cell death when added within 3 h after stimulation with anti-IgM Ab. **Treatment** with Abs against CD11a, CD18, or CD54 inhibited not only the homotypic adhesion but also the inhibition of anti-IgM Ab-induced apoptosis by anti-CD40 Ab. CD11a **antisense** decreased the surface CD11a expression, the anti-CD40 Ab-induced homotypic adhesion, and the inhibitory effect of anti-CD40 Ab on anti-IgM Ab-induced apoptosis. The data show that LFA-1/ICAM-1-dependent cell adhesion induced by signaling through CD40 plays an important role in the inhibition of anti-IgM Ab-induced apoptosis of DND-39 cells.

4/3,AB/91 (Item 91 from file: 155)
DIALOG(R)File 155:MEDLINE(R)
(c) format only 2001 Dialog Corporation. All rts. reserv.

08436105 94341744 PMID: 8063271
Meeting report: **antisense** oligonucleotides.
Martinelli G; Ferrari S
Istituto di Ematologia L.e A. Seragnoli, Universita di Bologna, Italy.
Haematologica (ITALY) Mar-Apr **1994**, 79 (2) p184-8, ISSN 0390-6078 Journal Code: FYB
Languages: ENGLISH
Document type: Congresses
Record type: Completed

The use of **antisense** oligonucleotides as a therapeutic tool in modulating gene expression represents a newly established strategy for **treating** diseases. Such oligomers may be designed to complement a region of a specific gene or messenger RNA. Using this approach, oligonucleotides can serve as a potential block of transcription or translation through sequence-specific hybridization with targeted genetic segments. In the Fourth Meeting of the Italian Society of Experimental Hematology "Discutiamone Insieme", authors reported the use of in vitro synthesized oligonucleotides to inhibit normal and chimeric gene expression of **bcl-2** in normal and neoplastic cell lines, respectively, that carry the t(14;18) translocation. The roles of c-myb and B-myb in the control of the proliferation and differentiation of normal hematopoietic cell lines have been investigated by selective inhibition of the expression of specific transcripts. To get some insight into the correlation between proliferation and differentiation in myeloid cells, some authors studied and reported the differentiation potential of G1-arrested cells obtained by a specific oligodeoxynucleotide complementary to the 5' region of the c-myb mRNA. The use of anti-P53 **antisense** oligos in the modulation of the growth of normal and neoplastic bone marrow progenitors was presented and confirmed the pivotal role of this gene in cell cycle control. The role of abl gene expression in normal and chronic myelogenous leukemia (CML) cells

is not yet completely understood. Selective inhibition of this proto-oncogene and of the abl-bcr oncogene have been achieved by using of c-abl sequence specific **antisense** oligonucleotides; this approach sheds new light on the function of this gene in CML.(ABSTRACT TRUNCATED AT 250 WORDS)

4/3,AB/92 (Item 92 from file: 155)
DIALOG(R)File 155:MEDLINE(R)
(c) format only 2001 Dialog Corporation. All rts. reserv.

08422924 94226946 PMID: 8172820
Regulation of chemoresistance by the **bcl-2** oncoprotein in non-Hodgkin's lymphoma and lymphocytic leukemia cell lines.
Reed JC; Kitada S; Takayama S; Miyashita T
La Jolla Cancer Research Foundation, Cancer Research Center, California.
Annals of oncology (NETHERLANDS) **1994**, 5 Suppl 1 p61-5, ISSN 0923-7534 Journal Code: AYF
Contract/Grant No.: CA-47956, CA, NCI; CA-60381, CA, NCI
Languages: ENGLISH
Document type: Journal Article
Record type: Completed
BACKGROUND: The **bcl-2** gene becomes activated by 14;18 chromosomal translocations in the majority of low-grade non-Hodgkin's lymphomas (NHLs) and is expressed at high levels in the absence of gene rearrangements in a high proportion of B-cell chronic lymphocytic leukemias (B-CLLs). The protein encoded by **bcl-2** contributes to neoplastic cell expansion by prolonging cell survival through its ability to block programmed cell death (apoptosis). Because many chemotherapeutic drugs have been shown ultimately to kill tumor cells through mechanisms consistent with programmed cell death, we tested whether the relative levels of **bcl-2** oncoprotein influence the sensitivity of lymphoma and leukemia cell lines to killing by conventional cytotoxic drugs commonly used in the **treatment** of cancer. METHODS: Leukemia cell lines with low levels of **bcl-2** expression were stably infected with recombinant **bcl-2** retroviruses to achieve elevations in **bcl-2** protein levels. Lymphoma cell lines with high levels of **bcl-2** expression as the result of 14;18 translocations were either stably transfected with inducible **bcl-2** **antisense** expression plasmids or **treated** with **bcl-2** **antisense** oligonucleotides to achieve reductions in **bcl-2** protein levels. The sensitivity of these genetically modified cells to killing by various antineoplastic drugs was then determined. RESULTS: Gene transfer-mediated elevations in **bcl-2** protein levels in lymphocytic leukemia cell lines was correlated with markedly elevated resistance to killing by all cytotoxic drugs tested. Conversely, **antisense**-mediated reductions in **bcl-2** protein levels in t(14;18)-containing NHL cell lines resulted in enhanced sensitivity to all anticancer drugs. CONCLUSIONS: The relative levels of **bcl-2** oncoprotein represent one of the key determinants of the sensitivity of lymphocytic cells to killing by essentially all drugs currently available for the **treatment** of cancer.

4/3,AB/93 (Item 93 from file: 155)
DIALOG(R)File 155:MEDLINE(R)
(c) format only 2001 Dialog Corporation. All rts. reserv.

08422907 95126512 PMID: 7825961
Anticancer drug resistance and inhibition of apoptosis.
Desoize B
GIBSA, Institut Godinot, Reims, France.
Anticancer research (GREECE) Nov-Dec **1994**, 14 (6A) p2291-4, ISSN 0250-7005 Journal Code: 59L
Languages: ENGLISH

Document type: Journal Article; Review; Review, Tutorial
Record type: Complet

Apoptosis is a new concept which could be of great importance in the understanding and **treatment** of cancer. An important feature is the discovery of inhibitors of apoptosis, because they induce resistance to chemotherapeutic drugs and irradiation. **Bcl-2** is the most well known of these apoptosis inhibitors. When it is overexpressed cells are less sensitive to cytotoxic drugs; on the contrary, when it is underexpressed they are more sensitive. Clinically, **bcl-2** expression is associated with a poor prognosis in several cancers. **Bcl-2** protein, p26-**bcl-2**, is located in the outer mitochondrial membrane, the nuclear envelope and the smooth endoplasmic reticulum. P26-**bcl-2** is an antioxidant; this property could explain the anti-apoptotic activity since peroxides seem to be important mediators of apoptosis. **Bcl-2 antisense** oligonucleotides are able to reverse the apoptosis inhibition. New cancer **treatments** should take into account the expression of **bcl-2**.

4/3,AB/94 (Item 94 from file: 155)
DIALOG(R)File 155:MEDLINE(R)
(c) format only 2001 Dialog Corporation. All rts. reserv.

08417172 94366757 PMID: 8084613

Antisense oligonucleotides suppress B-cell lymphoma growth in a SCID-hu mouse model.

Cotter FE; Johnson P; Hall P; Pocock C; al Mahdi N; Cowell JK; Morgan G
LRF Department of Haematology and Oncology, Institute of Child Health, London.

Oncogene (ENGLAND) Oct **1994**, 9 (10) p3049-55, ISSN 0950-9232
Journal Code: ONC
Languages: ENGLISH
Document type: Journal Article
Record type: Completed

The t(14;18) translocation is found in the majority follicular lymphomas and some high grade B-cell lymphomas. This is results in deregulation of the **BCL-2** gene and appears to play a role in oncogenesis. Various numbers of cells from a cell line derived spontaneously from a patient with B-cell lymphoma bearing the t(14;18) translocation and negative for the Epstein-Barr virus (EBV) were injected by IP, IV, and SC routes into SCID mice. The mice developed lymphoma bearing the t(14;18) translocation with as few as 5 x 10(6) cells within 28 days. This was determined by histological examination. The higher the cell inoculation the more rapidly the lymphoma developed. Engraftment of the tumour cells was determined by PCR for the t(14;18) breakpoint region on peripheral blood samples and could be detected prior to development of overt lymphoma. Having established a lymphoma model the cells were **treated** with **antisense** oligonucleotides to the first open reading frame of the **BCL-2** gene prior to inoculation of the SCID mice. Control **treatments** with sense and nonsense oligonucleotides was also performed. At 28 days the sense, nonsense and untreated cell SCID mice had developed lymphoma, however, the **antisense treated** group failed to develop lymphoma. The findings demonstrate the modelling of B-cell lymphoma bearing the t(14;18) translocation and the ability to modify the lymphoma process with the use of **antisense** oligonucleotides to the **BCL-2** gene. Reduction of the BCL2 protein suppresses the oncogenic potential of these lymphoma cells confirming that it plays an essential role in the development of malignancy.

4/3,AB/95 (Item 95 from file: 155)
DIALOG(R)File 155:MEDLINE(R)
(c) format only 2001 Dialog Corporation. All rts. reserv.

08412081 94297233 PMID: 8025285

Effects of **BCL-2 antisense** oligodeoxynucleotides on in vitro proliferation and survival of normal marrow progenitors and leukemic cells.

Campos L; Sabido O; Rouault JP; Guyotat D

Centre de Transfusion Sanguine, Lyon, France.

Blood (UNITED STATES) Jul 15 **1994**, 84 (2) p595-600, ISSN 0006-4971 Journal Code: A8G

Languages: ENGLISH

Document type: Journal Article

Record type: Completed

Previous studies have shown that the **BCL-2** protooncogene encodes a mitochondrial protein that promotes cell survival by blocking programmed cell death. **Bcl-2** protein has been detected in normal immature myeloid cells and in acute myeloid leukemia (AML) cells. To assess its functional role in normal and leukemic hematopoiesis, we performed serum-free cultures of CD34+ normal marrow cells, of **bcl-2**-positive myeloid lines, and of AML cells in the presence of **bcl-2** sense, nonsense, and **antisense** phosphorothioate oligodeoxynucleotides. In all **antisense-treated** cultures, we observed (1) an inhibition of **bcl-2** protein expression by day 4 to 6 of culture; (2) a decrease in cell survival duration; and (3) a decrease in the number of clonogenic cells present in the culture. Moreover, exposure to chemotherapeutic drugs resulted in more effective killing of AML cells in the presence of **antisense** oligomers. We conclude that **bcl-2** protein is necessary for the survival of myeloid cells in culture, and that it may be implicated in the resistance of AML cells to chemotherapy.

4/3,AB/96 (Item 96 from file: 155)

DIALOG(R)File 155:MEDLINE(R)

(c) format only 2001 Dialog Corporation. All rts. reserv.

08270644 95037634 PMID: 7950302

Reversal of chemoresistance of lymphoma cells by **antisense**-mediated reduction of **bcl-2** gene expression.

Kitada S; Takayama S; De Riel K; Tanaka S; Reed JC

La Jolla Cancer Research Foundation, Cancer Research Center, California 92037.

Antisense research and development (UNITED STATES) Summer **1994**, 4 (2) p71-9, ISSN 1050-5261 Journal Code: BI7

Contract/Grant No.: CA-60381, CA, NCI

Languages: ENGLISH

Document type: Journal Article

Record type: Completed

The **bcl-2** gene is expressed in many types of human tumours and becomes transcriptionally deregulated in the majority of non-Hodgkin's lymphomas as the result of t(14;18) chromosomal translocations. The 26-kDa **Bcl-2** protein has been shown to block programmed cell death (apoptosis) induced by many types of stimuli, including a wide variety of chemotherapeutic drugs and radiation. The presence of **bcl-2** in tumor cells has been correlated with poor responses to therapy in patients with some types of cancer. To explore further the relevance of **bcl-2** to drug resistance, we used **antisense** (As) approaches to achieve reductions in the levels of steady state **Bcl-2** protein levels in t(14;18)-containing human lymphoma cell lines. Both synthetic **bcl-2** -As oligonucleotides and inducible expression plasmids that produce **bcl-2**-As transcripts induced reductions in **bcl-2** expression, resulting in a marked enhancement in the sensitivity of neoplastic cells to conventional chemotherapeutic drugs such as cytosine arabinoside (ara-C) and methotrexate (MTX). These results suggest that novel therapeutics targeted against **bcl-2** could provide the means for improved **treatment** of cancer by affecting physiological pathways distal to the targets of cytotoxic drugs.

4/3,AB/97 (Item 97 from file: 155)
DIALOG(R)File 155:MEDLINE(R)
(c) format only 2001 Dialog Corporation. All rts. reserv.

08050526 94067789 PMID: 8247541
Apoptosis in Burkitt lymphoma cells is driven by c-myc.
Milner AE; Grand RJ; Waters CM; Gregory CD
Department of Immunology, University of Birmingham Medical School, UK.
Oncogene (ENGLAND) Dec **1993**, 8 (12) p3385-91, ISSN 0950-9232
Journal Code: ONC
Languages: ENGLISH
Document type: Journal Article
Record type: Completed
Chromosomal translocation and subsequent de-regulation of the c-myc proto-oncogene are considered to be critical events in the multi-stage evolution of Burkitt lymphoma (BL). It is widely accepted that Myc protein functions as a competence factor for proliferation. However, recent studies indicate that it can also act in some cell types as a regulator of apoptosis. BL cell populations display a high frequency of apoptosis in vivo, a property which is also readily demonstrable in vitro in group I BL cell lines. Such lines are known to retain the cell surface marker characteristics of the parental tumour cells and, in the case of Epstein-Barr virus-positive tumours, their restricted viral protein expression. We have shown previously that apoptosis in a group I BL cell line is inhibited by interferon (IFN)-alpha. Here we show that IFN-alpha-mediated suppression of apoptosis in group I BL cells corresponds temporally with inhibition of Myc protein levels. Furthermore, inhibition of Myc expression following **treatment** with c-myc anti-sense oligonucleotides markedly enhanced survival of group I BL cells. These results indicate that, whilst c-myc may facilitate cycling of tumour cells in which it is de-regulated, it also stimulates their apoptosis.

4/3,AB/98 (Item 98 from file: 155)
DIALOG(R)File 155:MEDLINE(R)
(c) format only 2001 Dialog Corporation. All rts. reserv.

08045398 94003939 PMID: 8400801
Investigations of **antisense** oligonucleotides targeted against **bcl-2** RNAs.
Kitada S; Miyashita T; Tanaka S; Reed JC
La Jolla Cancer Research Foundation, Cancer Research Center, California.
Antisense research and development (UNITED STATES) Summer **1993**, 3 (2) p157-69, ISSN 1050-5261 Journal Code: BI7
Contract/Grant No.: CA-60381, CA, NCI
Languages: ENGLISH
Document type: Journal Article
Record type: Completed
Expression of the **bcl-2** gene becomes deregulated in many non-Hodgkin lymphomas as the result of t(14;18) chromosomal translocations. Because **bcl-2** regulates the survival of cells, and because its over-expression is associated with cellular resistance to killing by chemotherapeutic drugs and gamma-irradiation, this gene and its mRNA and protein products represent ideal targets for designing novel therapeutic strategies for the **treatment** of cancer. Here we describe the effects of an 18-mer phosphodiester oligonucleotide that is complementary to the first 6 codons of the **bcl-2** mRNA's open reading frame. When tested for inhibition of in vitro protein synthesis using RNAse-H-supplemented reticulocyte lysates and RNA prepared by in vitro transcription of a human **bcl-2** cDNA, the **bcl-2** **antisense** (AS) oligomer completely abolished **Bcl-2** protein production at 10 microM, but had no effect on the in vitro translation of a chicken **bcl-2** RNA that contained three mismatches relative to the oligomer binding site on the human **bcl-**

2 RNA. A control 18-mer having the same base composition as the AS oligomer but with scrambled order (SC) was not inhibitory. Addition of AS and SC oligomers to cultures of a NIH-3T3 fibroblast cell line that had been stably infected with a recombinant retrovirus containing the same human **bcl-2** cDNA used for in vitro transcription/translation experiments revealed concentration-dependent reductions in the relative levels of the 26-kD human **Bcl-2** protein (as determined by immunoblotting) by the AS but not by the SC oligomer. Similar results were obtained when AS and SC oligomers were applied to a t(14;18)-containing lymphoma cell line SU-DHL-4 that was cultured in low-serum media. When used at 200 microM, the **bcl-2** AS oligomer produced 84-95% reductions in **Bcl-2** protein levels in SU-DHL-4 cells but had relatively little effect on the levels of other mitochondrial control proteins, suggesting that the inhibitory effects were specific. **Treatment** of SU-DHL-4 cells with AS oligomer lead to essentially complete loss of **bcl-2** mRNA from cells within 1 day of addition to cultures, but presumably because of the long half-life of the **Bcl-2** protein (approximately 14 h), commensurate reductions in **Bcl-2** protein levels did not occur until 3 days. (ABSTRACT TRUNCATED AT 400 WORDS)

4/3,AB/99 (Item 1 from file: 5)
DIALOG(R)File 5:Biosis Previews(R)
(c) 2001 BIOSIS. All rts. reserv.

12541162 BIOSIS NO.: 200000294664
Antisense transcript associated to tumor cells having a T(14;18) translocation and oligodeoxynucleotides useful in the diagnosis and **treatment** of said tumor cells.
AUTHOR: Capaccioli Sergio(a); Morelli Susann; Nicolin Angelo
AUTHOR ADDRESS: (a)Florence**Italy
JOURNAL: Official Gazette of the United States Patent and Trademark Office Patents 1229 (3):pNo pagination Dec. 21, **1999**
MEDIUM: e-file.
ISSN: 0098-1133
DOCUMENT TYPE: Patent
RECORD TYPE: Abstract
LANGUAGE: English

ABSTRACT: A chimeric **bcl-2**/IgH **antisense** transcript that hybridizes with the pre-mRNA of a hybrid gene in t(14;18) translocated cells. An ODN directed to complement any region of the above mentioned **antisense** transcript and the use thereof for diagnostic or therapeutic purposes.

1999

4/3,AB/100 (Item 2 from file: 5)
DIALOG(R)File 5:Biosis Previews(R)
(c) 2001 BIOSIS. All rts. reserv.

12524385 BIOSIS NO.: 200000277887
Antisense modulation of novel anti-apoptotic **bcl-2** -related proteins.
AUTHOR: Ackermann Elizabeth J; Bennett C Frank; Dean Nicholas M; Marcusson Eric G(a)
AUTHOR ADDRESS: (a)San Diego, CA**USA
JOURNAL: Official Gazette of the United States Patent and Trademark Office Patents 1229 (2):pNo pagination Dec. 14, **1999**
MEDIUM: e-file.
ISSN: 0098-1133
DOCUMENT TYPE: Patent
RECORD TYPE: Abstract
LANGUAGE: English

ABSTRACT: Compositions [redacted] methods are provided for modu[redacted]ing the expression of novel anti-apoptotic **bcl-2**-related proteins. **Antisense** oligonucleotides targeted to nucleic acids encoding the human novel anti-apoptotic **bcl-2**-related proteins A1 and mcl-1 are preferred. Methods of using these compounds for modulation of novel anti-apoptotic **bcl-2**-related protein expression and for **treatment** of diseases associated with expression of novel anti-apoptotic **bcl-2**-related proteins are also provided. Also provided are methods of using these compounds for promoting apoptosis and for **treatment** of diseases for which promotion of apoptosis is desired.

1999

4/3,AB/101 (Item 3 from file: 5)
DIALOG(R)File 5:Biosis Previews(R)
(c) 2001 BIOSIS. All rts. reserv.

12301874 BIOSIS NO.: 200000059741
Establishment of an apoptotic model of BGC-823 gastric cancer cell line with expression of **bcl-2** gene.
AUTHOR: Lu Ping(a); Chen Junqing(a); Wang Shubao(a)
AUTHOR ADDRESS: (a)The First Hospital of China Medical University, Shenyang **China
JOURNAL: Zhongguo Zhongliu Linchuang 26 (8):p586-588 Aug. 20, **1999**
ISSN: 1000-8179
DOCUMENT TYPE: Article
RECORD TYPE: Abstract
LANGUAGE: Chinese; Non-English
SUMMARY LANGUAGE: Chinese; English

ABSTRACT: Using phase and electronic microscopy, flow cytometry, electrophoresis of DNA the apoptosis of BGC-823 gastric cancer cell line was observed. The results showed that twenty-four hrs after being **treated** with **bcl-2 antisense** oligodeoxynucleotides (30uM), Cisplatin (2ug/ml), Carboplantin (20ug/ml), Adriamycin (4ug/ml), and Mitomycin (4ug/ml), the tumor cells shrinked with condensed chromatin, and apoptic bodies. Hypodiploid DNA was observed by flow cytometry and so did fragmentation of chromosomal DNA by electrophoresis of DNA.

1999

4/3,AB/102 (Item 4 from file: 5)
DIALOG(R)File 5:Biosis Previews(R)
(c) 2001 BIOSIS. All rts. reserv.

12259196 BIOSIS NO.: 200000012698
Effect of WT1 gene expression on cell growth and proliferation in myeloid leukemia cell lines.
AUTHOR: Mi Yingchang(a); Wang Li(a); Bian Shougeng(a); Meng Qingxiang(a); Chen Guibin(a); Wang Jianxiang(a)
AUTHOR ADDRESS: (a)Institute of Hematology and Blood Diseases Hospital, CAMS and PUMC, Tianjin**China
JOURNAL: Chinese Medical Journal (English Edition) 112 (8):p705-708 Aug., **1999**
ISSN: 0366-6999
DOCUMENT TYPE: Article
RECORD TYPE: Abstract
LANGUAGE: English
SUMMARY LANGUAGE: English

ABSTRACT: Objective: To investigate the effects and mechanism of Wilms' tumor (WT1) **antisense** oligonucleotides (AS-oligomers) on proliferation and apoptosis in meyloid leukemia cell lines. Methods: K562 and HL-60 cells were cultured in presence of WT1 oligomers. Both cell lines express WT1 gene with no p53 protein expression. Cells growth, apoptosis and expression of WT1, **bcl-2** genes were analysed using 3-(4, 5-dimethylthiazol-2-yl)-2, 5-diphenylmetrazolium bromide (MTT) colorimetric assay, flow cytometry and reverse transcription-polymerase chain reaction (RT-PCR) methods. Results: WT1 **antisense** oligonucleotides inhibited cellular proliferation of K562 cells and the effect was concentration-dependent. When cultured at concentration of 200 mug/ml oligomers, growth inhibition was 46.2% for **antisense** oligonucleotide cultivated group and 28.1% for sense oligonucleotide cultured group (P = 0.008) respectively. WT1 **antisense** oligonucleotide can induce apoptosis of K562 and HL-60 cells. Per centages of apoptotic cells in **antisense** oligonucleotide and sense oligonucleotide **treated** groups were 30.88% versus 13.62% for K562 cells and 40.15% versus 4.23% for HL-60 cells. However the growth of HL-60 cells and expression of **bcl-2** gene were unaffected. Conclusions: The WT1 gene is related with proliferation and apoptosis of leukemic cells. Effect of anti-apoptosis may be independent of the cellular p53 status and **bcl-2** expression. WT1 gene may play an important role in leukemogenesis.

1999

4/3,AB/103 (Item 5 from file: 5)
DIALOG(R)File 5:Biosis Previews(R)
(c) 2001 BIOSIS. All rts. reserv.

12259038 BIOSIS NO.: 200000012540
Apoptosis of human gastric cancer cells induced by **bcl-2** **antisense** oligodeoxynucleotides.
AUTHOR: Lu Ping(a); Chen Junqing(a)
AUTHOR ADDRESS: (a)The First Affiliated Hospital, China Medical University, Shenyang, 110001**China
JOURNAL: Zhonghua Zhongliu Zazhi 21 (4):p253-255 July, **1999**
ISSN: 0253-3758
DOCUMENT TYPE: Article
RECORD TYPE: Abstract
LANGUAGE: Chinese; Non-English
SUMMARY LANGUAGE: Chinese; English

ABSTRACT: Objective To study the regulation of **bcl-2** gene expression and induction of apoptosis by **bcl-2** **antisense** oligodeoxynucleotides (AS-ODN) on human gastric cancer cell line BGC-823 in vitro. Methods Two **bcl-2** AS-ODNs were synthesized, one covering the initiation sequence of translation of **bcl-2** mRNA (AS-ODN1) and the other covering the protein coding region (AS-ODN2). BGC-823 cells in logarithmic phase of growth were cultured in the presence of free or liposome (DOTAP)-encapsulated AS-ODN. Cell growth was assessed by MTT method. The expression of **bcl-2** at mRNA and protein levels was examined by RT-PCR and flow cytometry, respectively. Electron microscopy and flow cytometry were used to demonstrate apoptotic changes in AS-ODN-**treated** cells. Results Both AS-ODNs inhibited proliferation of BGC-823 cells. The inhibitory activity of AS-ODN2 was stronger than that of AS-ODN1. AS-ODNs encapsulated in liposome led to more marked inhibition of cell growth than free AS-ODNs. Both AS-ODNs reduced bc 1-2 expression of BGC-823 cells at mRNA and protein levels. Apoptosis of BGC-823 cells were demonstrated by the appearance of apoptotic bodies, chromatin condensation and pre-G1 peak on flow cytometric analysis. Conclusion **Antisense** oligodeoxynucleotide of **bcl-2** decreases **bcl-2** gene expression and induces apoptosis of human gastric

cancer cells in vitro

1999

4/3,AB/104 (Item 6 from file: 5)
DIALOG(R)File 5:Biosis Previews(R)
(c) 2001 BIOSIS. All rts. reserv.

12141741 BIOSIS NO.: 199900436590
Results of a phase I clinical trial of **bcl-2 antisense** molecule G3139 (Genta) in patients with non-Hodgkin's lymphoma (NHL).
AUTHOR: Waters J S(a); Webb A(a); Cunningham D(a); Clarke P A; di Stefano F ; Raynaud F; Brown B D; Cotter F
AUTHOR ADDRESS: (a)Royal Marsden Hospital, London**UK
JOURNAL: British Journal of Cancer 80 (SUPPL. 2):p62 July, **1999**
CONFERENCE/MEETING: Joint Meeting of the British Association for Cancer Research, the British Oncological Association, the Association of Cancer Physicians and the Royal College of Radiologists Edinburgh, Scotland, UK July 11-14, 1999
ISSN: 0007-0920
RECORD TYPE: Citation
LANGUAGE: English
1999

4/3,AB/105 (Item 7 from file: 5)
DIALOG(R)File 5:Biosis Previews(R)
(c) 2001 BIOSIS. All rts. reserv.

11993912 BIOSIS NO.: 199900274431
Selective activation of Ha-rasval12 oncogene increases susceptibility of NIH/3T3 cells to TNF-alpha.
AUTHOR: Chang Meng-Yao; Won Shen-Jeu; Yang Bei-Chang; Jan Ming-Shiou; Liu Hsiao-Sheng(a)
AUTHOR ADDRESS: (a)Department of Microbiology and Immunology, College of Medicine, National Cheng Kung University, **China
JOURNAL: Experimental Cell Research 248 (2):p589-598 May 1, **1999**
ISSN: 0014-4827
DOCUMENT TYPE: Article
RECORD TYPE: Abstract
LANGUAGE: English
SUMMARY LANGUAGE: English

ABSTRACT: This is the first report demonstrating that NIH/3T3 fibroblasts utilize the Raf-1/MAPK pathway to sensitize themselves to tumor necrosis factor-alpha (TNF-alpha) cytotoxicity under Ha-rasVal12 oncogene-overexpressed conditions. This paper clearly shows that the sensitivity of NIH/3T3 cells to TNF-alpha cytotoxicity positively correlated with the expression level of activated Ha-ras transgene, which was manipulated either positively by isopropyl-beta-D-thiogalactoside (IPTG) induction or negatively by a **ribozyme** or a dominant negative Ras suppression. Further analysis revealed that after TNF-alpha **treatment**, Ha-ras-overexpressed transformants underwent apoptosis. Overexpression of dominant negative Raf-1, Rac1, or RhoA in the Ha-ras transformants clarified that among these factors, only dominant negative Raf-1 could reverse the cell sensitivity to TNF-alpha, indicating that Raf-1, as a proapoptotic factor, indeed participates in TNF-alpha cytotoxicity. The anti-apoptotic roles of **Bcl-2** and PI(3) kinase are also demonstrated by the Ha-ras transformants which became more resistant to TNF-alpha while overexpressing **Bcl-2** or the activated p110 catalytic subunit. The analyses of the cell cycle and nuclear transcription factor activities revealed that TNF-alpha **treatment** caused the Ha-ras overexpressed transformants to shift from S to G0/G1 phase and increased the responses of AP-1, c-fos, and

c-myc. Taken together, we suggest that the possible action of Ha-ras overexpression to sen[redacted]ize TNF-alpha-**treated** fibrobla[redacted] is predominantly through the Ras/Raf-1/MAPK pathway to in[redacted]ease the responses of AP-1, c-fos, and c-myc, which are possibly involved in the aberration of cell cycle machinery, and subsequently to turn on the death program.

1999

4/3,AB/106 (Item 8 from file: 5)
DIALOG(R)File 5:Biosis Previews(R)
(c) 2001 BIOSIS. All rts. reserv.

11968624 BIOSIS NO.: 199900221937
Bcl-2 antisense therapy in multiple myeloma.
AUTHOR: Bloem A(a); Lockhorst H
AUTHOR ADDRESS: (a)Department of Immunology, University Hospital Utrecht, Heidelberglaan 100, 3584 CX, Utrecht**Netherlands
JOURNAL: Pathologie Biologie 47 (2):p216-220 Feb., **1999**
ISSN: 0369-8114
DOCUMENT TYPE: Literature Review
RECORD TYPE: Abstract
LANGUAGE: English
SUMMARY LANGUAGE: English; French

ABSTRACT: Multiple myeloma is a plasma cell tumor localised in the bone marrow. During chemotherapy drug resistance develops in almost all patients. We have indications that the anti-apoptopic protein **Bcl-2** is a key element in multi drug resistance in myeloma. Reduction of **Bcl-2** renders the tumor cell susceptible to drug induced apoptosis. This suggests that therapies directed at lowering **Bcl-2** levels in myeloma cells in vivo, like **Bcl-2 antisense treatment**, might chemosensitize the tumor cells and therefore might be applicable for therapeutic use.

1999

4/3,AB/107 (Item 9 from file: 5)
DIALOG(R)File 5:Biosis Previews(R)
(c) 2001 BIOSIS. All rts. reserv.

11957480 BIOSIS NO.: 199900203589
Hyperoxia induces the neuronal differentiated phenotype of PC12 cells via a sustained activity of mitogen-activated protein kinase induced by **Bcl-2**.
AUTHOR: Katoh Shinsuke; Mitsui Youji; Kitani Kenichi; Suzuki Takahiko(a)
AUTHOR ADDRESS: (a)Faculty of Medicine, Radiation Research Institute, University of Tokyo, 7-3-1, Hongo, Bunkyo-ku,**Japan
JOURNAL: Biochemical Journal 338 (2):p465-470 March 1, **1999**
ISSN: 0264-6021
DOCUMENT TYPE: Article
RECORD TYPE: Abstract
LANGUAGE: English
SUMMARY LANGUAGE: English

ABSTRACT: We previously reported that rat pheochromocytoma PC12 cells express the neuronal differentiated phenotype under hyperoxia through the production of reactive oxygen species (ROS). In the present study, we found that in this phenotype, **Bcl-2**, an apoptosis inhibitor, affects mitogen-activated protein (MAP)-kinase activity, which is known as a key enzyme of the signal-transduction cascade for differentiation. When PC12 cells were cultured under hyperoxia, a rapid increase in MAP-kinase activity, including that of both p42 and p44, was observed.

Although the activity level then decreased quickly, activity higher than the control level was observed for 48 h. PD98059, an inhibitor of MAP kinase, suppressed the hyperoxia-induced neurite extensions, suggesting the involvement of MAP-kinase activity in the mechanism of differentiation induced by ROS. An elevation of **Bcl-2** expression was observed after culturing PC12 cells for 24 h under hyperoxia. This **Bcl-2** elevation was not affected by **treatment** with PD98059, suggesting that it did not directly induce neurite extension under hyperoxia. However, the blockade of the **Bcl -2** elevation by an **antisense** oligonucleotide inhibited the sustained MAP-kinase activity and neurite extensions under hyperoxia. Further, in PC12 cells highly expressing **Bcl-2**, the sustained MAP-kinase activity and neurite extensions under hyperoxia were enhanced. These results suggested that MAP kinase is activated through the production of ROS, and the subsequent elevation of **Bcl-2** expression sustains the MAP-kinase activity, resulting in the induction of the neuronal-differentiation phenotype of PC12 cells under hyperoxia.

1999

4/3,AB/108 (Item 10 from file: 5)
DIALOG(R)File 5:Biosis Previews(R)
(c) 2001 BIOSIS. All rts. reserv.

11938905 BIOSIS NO.: 199900185014
Downregulation of **bcl-2** expression by **antisense** -oligonucleotide (AS-ODN) **treatment** enhances mitoxantrone cytotoxicity in the androgen-dependent Shionogi tumor model.
AUTHOR: Tolcher A(a); Miyake H; Gleave M
AUTHOR ADDRESS: (a)Div. Urology, Vancouver Hosp., Vancouver, BC**Canada
JOURNAL: Proceedings of the American Association for Cancer Research Annual Meeting 40p484 March, **1999**
CONFERENCE/MEETING: 90th Annual Meeting of the American Association for Cancer Research Philadelphia, Pennsylvania, USA April 10-14, 1999
SPONSOR: American Association for Cancer Research
ISSN: 0197-016X
RECORD TYPE: Citation
LANGUAGE: English
1999

4/3,AB/109 (Item 11 from file: 5)
DIALOG(R)File 5:Biosis Previews(R)
(c) 2001 BIOSIS. All rts. reserv.

11935678 BIOSIS NO.: 199900181787
Antisense BCL-2 oligonucleotide enhances the chemosensitivity of taxol in the Shionogi tumor model.
AUTHOR: Miyake Hideaki; Tolcer Anthony; Gleave Martin
AUTHOR ADDRESS: Vancouver**Canada
JOURNAL: Journal of Urology 161 (4 SUPPL.):p130 April, **1999**
CONFERENCE/MEETING: 94th Annual Meeting of the American Urological Association, Inc. Dallas, Texas, USA May 1-6, 1999
SPONSOR: American Urological Association
ISSN: 0022-5347
RECORD TYPE: Citation
LANGUAGE: English
1999

4/3,AB/110 (Item 12 from file: 5)
DIALOG(R)File 5:Biosis Previews(R)
(c) 2001 BIOSIS. All rts. reserv.

11799674 BIOSIS NO.: 199900045783
BCL-2 antisense treatme[redacted] blocks induced tolerance to focal cerebral ischemia in the rat.
AUTHOR: Simon R P; Shigetoshi S; Zhu R; Graham S H; Henshall D C; Goss J R
AUTHOR ADDRESS: Dep. Neurol., Univ. Pittsburgh, Biomedical Science Tower S5, Pittsburgh, PA 15213**USA
JOURNAL: Society for Neuroscience Abstracts 24 (1-2):p253 **1998**
CONFERENCE/MEETING: 28th Annual Meeting of the Society for Neuroscience, Part 1 Los Angeles, California, USA November 7-12, 1998
SPONSOR: Society for Neuroscience
ISSN: 0190-5295
RECORD TYPE: Citation
LANGUAGE: English
1998

4/3,AB/111 (Item 13 from file: 5)
DIALOG(R)File 5:Biosis Previews(R)
(c) 2001 BIOSIS. All rts. reserv.

11731999 BIOSIS NO.: 199800513730
Growth factors prevent changes in **Bcl-2** and Bax expression and neuronal apoptosis induced by nitric oxide.
AUTHOR: Tamatani Michio(a); Ogawa Satoshi; Nunez Gabriel; Tohyama Masaya
AUTHOR ADDRESS: (a)Dep. Anat. Neuroscience, Osaka Univ. Med. Sch., 2-2 Yamadaoka, Suita, Osaka 565**Japan
JOURNAL: Cell Death and Differentiation 5 (10):p911-919 Oct., **1998**
ISSN: 1350-9047
DOCUMENT TYPE: Article
RECORD TYPE: Abstract
LANGUAGE: English

ABSTRACT: Recent studies have shown that nitric oxide (NO) donors can trigger apoptosis of neurons, and growth factors such as insulin-like growth factor-1 (IGF-1) and basic fibroblast growth factor (bFGF) can protect against NO-induced neuronal cell death. The purpose of this study was to elucidate the possible mechanisms of NO-mediated neuronal apoptosis and the neuroprotective action of these growth factors. Both IGF-1 and bFGF prevented apoptosis induced by NO donors, sodium nitroprusside (SNP) or 3-morpholinosydnonimin (SIN-1) in hippocampal neuronal cultures. Incubation of neurons with SNP induced caspase-3-like activation following downregulation of **Bcl-2** and upregulation of Bax protein levels in cultured neurons. **Treatment** of neurons with a bax **antisense** oligonucleotide inhibited the caspase-3-like activation and neuronal death induced by SNP. In addition, **treatment** of neurons with an inhibitor of caspase-3, Ac-DEVD-CHO, together with SNP did not affect the changes in the protein levels, although it inhibited NO-induced cell death. Pretreatment of cultures with either IGF-1 or bFGF prior to NO exposure Inhibited caspase-3-like activation together with the changes in **Bcl-2** and Bax protein levels. These results suggest that the changes in **Bcl-2** and Bax protein levels followed by caspase-3-like activation are a component In the cascade of NO-induced neuronal apoptosis, and that the neuroprotective actions of IGF-1 and bFGF might be due to inhibition of the changes in the protein levels of the **Bcl-2** family.

1998

4/3,AB/112 (Item 14 from file: 5)
DIALOG(R)File 5:Biosis Previews(R)
(c) 2001 BIOSIS. All rts. reserv.

11426775 BIOSIS NO.: 199800208107
Bcl-2-independent Bcr-Abl-mediated resistance to apoptosis:

Protection is correlated with up regulation of Bcl-x|L.
AUTHOR: Amarante-Mendes [redacted]stavo P(a); McGahon Anne J; Ni[redacted]oka Walter K; Afar Daniel E H; Witte Owen N; Green Douglas R
AUTHOR ADDRESS: (a)Dep. Imunol., Inst. Cienc. Biomed., Univ. Sao Paulo, Sao Paulo 05508-900**Brazil
JOURNAL: Oncogene 16 (11):p1383-1390 March 19, **1998**
ISSN: 0950-9232
DOCUMENT TYPE: Article
RECORD TYPE: Abstract
LANGUAGE: English

ABSTRACT: Bcr-Abl is the molecule responsible for both the transformation phenotype and the resistance to chemotherapeutic drugs found in chronic myelogenous leukemia (CML) cells. Wild-type HL-60, a transformed pro-myelocytic cell line, is very susceptible to apoptosis-inducing agents. We show here that expression of Bcr-Abl in HL-60 cells rendered them extremely resistant to apoptosis induced by a wide variety of agents. The anti-apoptotic effect of Bcr-Abl was found to be independent of the phase of the cell cycle. **Treatment** with **antisense** oligonucleotides directed to bcr decreased the expression of the ectopic bcr-abl and restored susceptibility to apoptosis. Double mutations affecting the autophosphorylation site and the phosphotyrosine-binding motif (FLVRES) have been previously shown to impair the transforming activity of Bcr-Abl in fibroblasts and hematopoietic cells, however HL-60 cells expressing this double mutant molecule exhibited the same level of resistance to apoptosis as those expressing the wild-type Bcr-Abl. Interestingly, wild type and mutant Bcr-Abl induced in HL-60 cells a dramatic down regulation of **Bcl-2** and increased the levels of Bcl-x|L. The level of Bax did not change in response to the presence of Bcr-Abl. **Antisense** oligonucleotides targeted to bcl-x down-regulated the expression of Bcl-x, and increased the susceptibility of HL-60.Bcr-Abl cells to staurosporine. Importantly, HL-60 cells overexpressing Bcl-x|L showed higher expression of Bcl-X|L but lower resistance to apoptosis when compared to HL-60.Bcr-Abl cells. The results described here show that Bcr-Abl is a powerful mammalian anti-apoptotic molecule and can act independently of **Bcl-2**. Bcl-X|L, however, seems to participate in part in Bcr-Abl-mediated resistance to apoptosis in HL-60 cells.

1998

4/3,AB/113 (Item 15 from file: 5)
DIALOG(R)File 5:Biosis Previews(R)
(c) 2001 BIOSIS. All rts. reserv.

11415378 BIOSIS NO.: 199800196710
Bcl-2 antisense oligodeoxynucleotide 2009 synergizes with chemotherapy on lung cancer cell lines and has antitumor activity against lung cancer xenografts.
AUTHOR: Zangemeister-Wittke U(a); Fabbro D; Mueller M; Schenker T; Stahel R A
AUTHOR ADDRESS: (a)Div. Oncol., Dep. Intern. Med., Univ. Hosp. Zurich, CH-8044 Zurich**Switzerland
JOURNAL: Proceedings of the American Association for Cancer Research Annual Meeting 39p417 March, **1998**
CONFERENCE/MEETING: 89th Annual Meeting of the American Association for Cancer Research New Orleans, Louisiana, USA March 28-April 1, 1998
SPONSOR: American Association for Cancer Research
ISSN: 0197-016X
RECORD TYPE: Citation
LANGUAGE: English
1998

4/3,AB/114 (Item 16 from file: 5)
DIALOG(R)File 5:Biosis Previews(R)
(c) 2001 BIOSIS. All rts. reserv.

11164090 BIOSIS NO.: 199799785235
Protein kinase C-beta-II activation by 1-beta-D-arabinofuranosylcytosine is antagonistic to stimulation of apoptosis and **Bcl-2**-alpha down-regulation.
AUTHOR: Whitman Susan P; Civoli Francesca; Daniel Larry W(a)
AUTHOR ADDRESS: (a)Dep. Biochem., Bowman Gray Sch. Med., Wake Forest Univ., Medical Center Blvd., Winston-Salem, NC**USA
JOURNAL: Journal of Biological Chemistry 272 (38):p23481-23484 **1997**
ISSN: 0021-9258
RECORD TYPE: Abstract
LANGUAGE: English

ABSTRACT: 1-beta-D-Arabinofuranosylcytosine (ara-C) stimulates the formation of both diglyceride and ceramide in the acute myelogenous leukemia cell line HL-60 (Strum, J. C., Small, G. W., Pauig, S. B., and Daniel, L. W. (1994) J. Biol. Chem 269, 15493-15497). ara-C also causes apoptosis in HL-60 cells which can be mimicked by exogenous ceramide. However, the signaling role for ara-C-induced diacylglycerol (DAG) is not defined. We found that **Bcl-2** levels were increased by **treatment** of HL-60 cells with exogenous DAG or 12-O-tetradecanoylphorbol-13-acetate (TPA). In contrast, exogenous ceramide **treatment** caused a decrease in cellular **Bcl-2** levels. Thus, ara-C stimulates the synthesis of two second messengers with opposing effects on **Bcl-2**. Since the effects of ara-C-induced DAG could be due to protein kinase C (PKC) activation, we determined the effects of ara-C on PKC isozymes. ara-C caused an increase in membrane-bound PKC-beta-II (but not PKC-alpha or PKC-delta). ara-C or TPA-induced translocation of PKC-beta-II was inhibited by 1-O-octadecyl-2-O-methyl-rac-glycero-3-phosphocholine (ET-18-OCH-3), and ara-C-induced apoptosis was stimulated by pretreatment of the cells with ET-18-OCH-3. ET-18-OCH-3 also inhibited stimulation of **Bcl-2** by TPA and enhanced the decrease in **Bcl-2** observed in ara-C-**treated** cells. These data indicate that ara-C-induced apoptosis is limited by ara-C-stimulated PKC-beta-II through effects on **Bcl-2**. To further determine the role of PKC, we used **antisense** oligonucleotides directed toward PKC-beta-II. The **antisense**, but not the sense, oligonucleotide inhibited PKC-beta-II activation and enhanced ara-C-induced apoptosis. These data demonstrate that the stimulation of apoptosis by ara-C is self-limiting and can be enhanced by inhibition of PKC.

1997

4/3,AB/115 (Item 17 from file: 5)
DIALOG(R)File 5:Biosis Previews(R)
(c) 2001 BIOSIS. All rts. reserv.

11058313 BIOSIS NO.: 199799679458
First demonstration of anti-lymphoma activity of **BCL-2** **antisense** molecule-G3139: Results of phase I/IIA clinical trial.
AUTHOR: Webb A(a); Cunningham D(a); Cotter F; Ross P(a); Walters J; Judson I(a); Raynaud F(a); Clarke P(a); Dziewanowska Z E
AUTHOR ADDRESS: (a)Royal Marsden Hosp., Sutton, Surrey**UK
JOURNAL: British Journal of Cancer 76 (SUPPL. 1):p33 **1997**
CONFERENCE/MEETING: Joint Meeting of the British Oncological Association, Association of Cancer Physicians and the Royal College of Radiologists St. Andrews, Scotland, UK July 5-8, 1997
ISSN: 0007-0920
RECORD TYPE: Citation
LANGUAGE: English

4/3,AB/116 (Item 18 from file: 5)
DIALOG(R)File 5:Biosis Previews(R)
(c) 2001 BIOSIS. All rts. reserv.

10899777 BIOSIS NO.: 199799520922
Effect of **antisense** oligodeoxynucleotide targeted against **bcl-2** gene on growth and apoptotic susceptibility of leukemia cells.
AUTHOR: Chen Xie-Qun Huang Gao-Sheng; Yang Ping-Di
AUTHOR ADDRESS: Dep. Hematol., Xijing Hosp., Fourth Military Med. Univ., Xi'an 710032**China
JOURNAL: Zhongguo Zhongliu Linchuang 24 (1):p9-12 **1997**
ISSN: 1000-8179
RECORD TYPE: Abstract
LANGUAGE: Chinese; Non-English
SUMMARY LANGUAGE: Chinese; English

ABSTRACT: After **treated** with **antisense** oligodeoxynucleotide specific for **bcl-2** gene for 3 days, the intrinsic **bcl-2** protein of T-lymphocytic leukemia cell line CEM reduced approximately by 50%, which caused target cells to decrease in survival and to be more sensitive to etoposide-induced apoptosis. The data presented here indicates that cellular intrinsic **bcl-2** protein may play an important role in the leukemic cells death triggered by apoptosis induced chemotherapeutic agents.
1997

4/3,AB/117 (Item 19 from file: 5)
DIALOG(R)File 5:Biosis Previews(R)
(c) 2001 BIOSIS. All rts. reserv.

10707340 BIOSIS NO.: 199799328485
Arsenic-induced neural tube defects in mice: Alterations in cell cycle gene expression.
AUTHOR: Wlodarczyk Bogdan J; Bennett Gregory D; Calvin Jim A; Finnell Richard H(a)
AUTHOR ADDRESS: (a)Dep. Veterinary Anat. Public Health, Texas A and Univ., College Station, TX 77843-4458**USA
JOURNAL: Reproductive Toxicology 10 (6):p447-454 **1996**
ISSN: 0890-6238
RECORD TYPE: Abstract
LANGUAGE: English

ABSTRACT: The potential of arsenic to cause neural tube defects (NTD) in the human population remains a topic of controversy. While clearly toxic, the lack of well-defined human epidemiologic studies on this subject has made it difficult to fully understand the effects arsenic may have on the developing human neural tube. In the absence of good clinical data, we have tried to develop a murine model where hypotheses about the reproductive toxicity of arsenate can be tested. For these studies a murine strain (LN/Bc) that has proven to be susceptible to arsenic-induced NTD was use. Because cellular proliferation is vital for normal neural tube closure (NTC) to occur, in the present study we investigated whether an acute arsenate **treatment** could alter the expression of several cell cycle genes during murine neurulation. Pregnant LM/Bc dams were injected intraperitoneally on gestation day (GD) 7:12 (day:hour) and 8:12 with 40 mg/kg of arsenate, a **treatment** that causes exencephaly in 90 to 100% of the exposed fetuses. Neural tubes were then isolated from both control and arsenic **treated** embryos at GD 9:00, 9:12, 10:00, and 10:12, which encompasses all the stages of neurulation for this murine strain. Using the molecular techniques of in situ transcription and **antisense** RNA amplification

(RT/aRNA) the expression pattern for **bcl-2**, p53, wee-1, and wnt-1 was analyzed at each of these time points. In the neural tubes isolated from control embryos, the expression of all four genes was significantly altered as neurulation progressed, demonstrating their developmental regulation. Following arsenate **treatment**, however, there was a significant upregulation in the expression of **bcl-2** and p53 at gestational day 9:0, compared to their control values. The heightened expression of both of these genes suggests that arsenic inhibits cell proliferation, rather than inducing apoptosis, which delayed NTC and ultimately led to the neural tube defects observed in exposed embryos.
1996

4/3,AB/118 (Item 20 from file: 5)
DIALOG(R)File 5:Biosis Previews(R)
(c) 2001 BIOSIS. All rts. reserv.

10449160 BIOSIS NO.: 199699070305
Inhibition of **bcl-2** protein expression by **antisense** S-oligodeoxynucleotides **treatment** exacerbates neuronal death after cerebral ischemia in rats.
AUTHOR: Chen J; Zhu R; Basta K; Simon R P; Graham S H
AUTHOR ADDRESS: Pittsburgh, PA**USA
JOURNAL: Neurology 46 (2 SUPPL.):pA270-A271 **1996**
CONFERENCE/MEETING: 48th Annual Meeting of the American Academy of Neurology San Francisco, California, USA March 23-30, 1996
ISSN: 0028-3878
RECORD TYPE: Citation
LANGUAGE: English
1996

4/3,AB/119 (Item 21 from file: 5)
DIALOG(R)File 5:Biosis Previews(R)
(c) 2001 BIOSIS. All rts. reserv.

10372653 BIOSIS NO.: 199698827571
Screening studies on expression of oncogenes in SRS lymphoma cell lines.
AUTHOR: Zheng Songguo Yin Lianhua(a); Xu Liangzhong(a); Ye Ming; Lu Biao; Zhu Zhendong(a)
AUTHOR ADDRESS: (a)Lab. Pathology, Cancer Hosp., Sch. Basic Med. Sci., Shanghai Med. Univ., Shanghai 200032**China
JOURNAL: Acta Academiae Medicinae Shanghai 23 (1):p7-9 **1996**
ISSN: 0257-8131
DOCUMENT TYPE: Article
RECORD TYPE: Abstract
LANGUAGE: Chinese; Non-English
SUMMARY LANGUAGE: Chinese; English

ABSTRACT: PURPOSE: The studies of oncogenes expression in a SRS - 82 mouse Lymphoma cell line and SAC - II B-2, SAC - II C-3 clones have been less reported. We must establish a oncogene spectrum for finishing the experimental **antisense treatment** to SRS lymphoma. METHODS: SRS - 82 mouse lymphoma cell line and SAC - II B-2, SAC - II C-3 clones were obtained from the Department of Pathophysiology, Shanghai Medical University. ABC immunohistochemical method was used. RESULTS: Strong staining was found for c - fos and c - myc, medium staining for c - jun, ras - p21 and c - erbB - 2, and negative reactions for P53 and **bcl - 2** in SRS - 82 cell line and its clones. Cell surface marks (CD-4 and CD-8) of these two clones and their parent cell line were negative, all of them belong to primary stem cell origin. CONCLUSIONS: Establishments of oncogene spectrum play an important role in the experimental **antisense treatment** in SRS lymphoma, and c - fos and c - myc were the best targets for the **antisense treatment**

1996

4/3,AB/120 (Item 22 from file: 5)
DIALOG(R)File 5:Biosis Previews(R)
(c) 2001 BIOSIS. All rts. reserv.

10358114 BIOSIS NO.: 199698813032
Preclinical pharmacokinetics of G3139, a phosphorothioate **antisense** to **bcl-2** in mice.
AUTHOR: Raynaud F(a); Orr R(a); Goddard P; Dizik M; Beck T; Vaghefi M; Woodle M; Judson I(a); Cotter F
AUTHOR ADDRESS: (a)CRC Cent. Therapeutics, The Inst. Cancer Res., 15 Cotswold Road, Sutton, Surrey**UK
JOURNAL: Proceedings of the American Association for Cancer Research Annual Meeting 37 (0):p411 **1996**
CONFERENCE/MEETING: 87th Annual Meeting of the American Association for Cancer Research Washington, D.C., USA April 20-24, 1996
ISSN: 0197-016X
RECORD TYPE: Citation
LANGUAGE: English
1996

4/3,AB/121 (Item 23 from file: 5)
DIALOG(R)File 5:Biosis Previews(R)
(c) 2001 BIOSIS. All rts. reserv.

10110203 BIOSIS NO.: 199698565121
Direct evidence for the participation of **bcl-2** in the regulation by retinoic acid of the ara-c sensitivity of leukemic stem cells.
AUTHOR: Hu Z-B; Minden M D; McCulloch E A(a)
AUTHOR ADDRESS: (a)Ontario Cancer Inst., 500 Sherbourne St. Toronto, ON M4X 1K9**Canada
JOURNAL: Leukemia (Basingstoke) 9 (10):p1667-1673 **1995**
ISSN: 0887-6924
DOCUMENT TYPE: Article
RECORD TYPE: Abstract
LANGUAGE: English

ABSTRACT: All-trans retinoic acid (ATRA) increases the sensitivity of AML blast cells to cytosine arabinoside (Ara-C) or daunorubicin (DNR) when ATRA is given after drug. We have proposed that down-regulation of **bcl-2** is part of the mechanism by which ATRA regulates drug sensitivity. To test this hypothesis cDNA encoding **bcl-2** was transfected into cells of the continuous lines OCI/AML-2 and OCI/AML-5. Four transfectant lines were isolated; three contained transfected **bcl-2** in the sense orientation (AML5-BCL2sa, AML5-BCL2sb and 2-bcl2) and one with anti-sense **bcl-2**(AML5-bcl2as). The presence of the transfected gene was demonstrated by Northern blot; translation of the sense transfected genes into protein was demonstrated by Western blotting. Lines with sense-oriented transfected **bcl-2** were significantly less sensitive to Ara-C or H-2O-2 than the parental lines; the cells with anti-sense transfected genes were more sensitive than their parent but the difference did not reach statistical significance. The effect of ATRA on **bcl-2** expression was compared in sense-transfected cells and their parents; by Northern blotting it was shown that the endogenous but not the transfected genes were down-regulated after ATRA exposure. The capacity of cells with transfected genes to respond to ATRA was tested by obtaining Ara-C survival curves for ATRA-**treated** cells. Compared to controls not exposed to ATRA, the transfected cells showed little or statistically

insignificant changes in Ara-C sensitivity after ATRA **treatment**. We conclude that data from the transfectants provides evidence that expression of **bcl-2** is a determinant of sensitivity to Ara-C and H-20-2; and that the effect of ATRA on sensitivity requires the presence of **bcl-2** genes in association with regulatory elements.

1995

4/3,AB/122 (Item 24 from file: 5)
DIALOG(R)File 5:Biosis Previews(R)
(c) 2001 BIOSIS. All rts. reserv.

09616275 BIOSIS NO.: 199598071193
In vivo engraftment and **BCL-2 antisense treatment** of low grade B-cell lymphoma lymph node biopsies in SCID mice.
AUTHOR: Cotter F; Hill M; Pocock C; Clarke P; Malone M; Cunningham D
AUTHOR ADDRESS: Dep. Haematol., Inst. Child Health, 30 Guilford St., London WC1 N1EH**UK
JOURNAL: Blood 84 (10 SUPPL. 1):p640A **1994**
CONFERENCE/MEETING: Abstracts Submitted to the 36th Annual Meeting of the American Society of Hematology Nashville, Tennessee, USA December 2-6, 1994
ISSN: 0006-4971
RECORD TYPE: Citation
LANGUAGE: English
1994

4/3,AB/123 (Item 1 from file: 42)
DIALOG(R)File 42:Pharmaceuticl News Idx
(c)2001 ProQuest Info&Learning. All rts. reserv.

00675341 48852142
G3139 on the fast track
AUTHOR: Anonymous
Applied Genetics News, v20, n4, p6
November 1, 1999
CODEN: AGNEEN DOCUMENT TYPE: Periodical; News JOURNAL CODE: AGN
LANGUAGE: English RECORD TYPE: Citation

4/3,AB/124 (Item 2 from file: 42)
DIALOG(R)File 42:Pharmaceuticl News Idx
(c)2001 ProQuest Info&Learning. All rts. reserv.

00647635 0647635
Antisense treats prostate cancer
Applied Genetics News, v19, n6, p3
Jan 1999
CODEN: AGNEEN JOURNAL CODE: AGN
LANGUAGE: English RECORD TYPE: Citation

4/3,AB/125 (Item 1 from file: 72)
DIALOG(R)File 72:EMBASE
(c) 2001 Elsevier Science B.V. All rts. reserv.

07931312 EMBASE No: 1999404640
Apoptosis regulating proteins as targets of therapy for haematological malignancies
Kornblau S.M.; Konopleva M.; Andreeff M.
S.M. Kornblau, Section Mol. Hematology and Therapy, MD Anderson Cancer Center, 1515 Holcombe Blvd., Houston, TX 77030-4095 United States

AUTHOR EMAIL: skornbl[redacted]@mdacc.tmc.edu
Expert Opinion on Inv[redacted]igational Drugs (EXPERT OPIN[redacted]VEST. DRUGS) (United Kingdom) 1999, 8/12 (2027-2057)
CODEN: EOIDE ISSN: 1354-3784
DOCUMENT TYPE: Journal; Review
LANGUAGE: ENGLISH SUMMARY LANGUAGE: ENGLISH
NUMBER OF REFERENCES: 306

Most chemotherapeutic agents used in the **treatment** of haematological malignancies cause cell death by inducing apoptosis through undefined means. The discovery of the proteins involved in apoptosis and the description of apoptotic pathways suggest new potential targets for therapeutic intervention. Both 'intrinsic' and 'extrinsic' pathways can be activated separately, but activation of caspases appears central to most apoptotic pathways. Novel approaches attempt to induce apoptosis by directly targeting a portion of an apoptotic pathway. Agents that trigger signalling of Fas or tumour necrosis factor- (TNF-) related apoptosis inducing ligand (TRAIL) receptor seek to induce the extrinsic pathway at the cell surface. The **BCL-2** family of proteins seems central to the regulation of those apoptotic pathways that involve mitochondrial sequestration or the release of cytochrome c, with subsequent activation of Apaf-1, caspase-9 and caspase-3. The activity of this family may depend upon both the phosphorylation state of different members and the relative level of pro- and anti-apoptotic members. New agents such as the staurosporine analogue UCN-01 and bryostatin are drought to affect apoptosis induction by altering **BCL-2** phosphorylation. Others, such as **BCL-2 antisense** and ATRA attempt tu modulate the protein levels to promote apoptosis. Direct activation of caspase-3 is a probable target, but as yet no agent with this direct function is in trial. Clinical trials of several agents have been completed or are underway. It is likely that agents that target particular points in apoptosis pathways will have antileukaemia/lymphoma activity, however, the optimal utilisation may involve combination with other more conventional agents that also activate apoptosis.

4/3,AB/126 (Item 2 from file: 72)
DIALOG(R)File 72:EMBASE
(c) 2001 Elsevier Science B.V. All rts. reserv.

07905936 EMBASE No: 1999379594
Role of CD14 expression in the differentiation-apoptosis switch in human monocytic leukemia cells **treated** with 1alpha,25-dihydroxyvitamin Dinf 3 or dexamethasone in the presence of transforming growth factor beta
Kanatani Y.; Kasukabe T.; Okabe-Kado J.; Yamamoto-Yamaguchi Y.; Nagata N.; Motoyoshi K.; Honma Y.
Y. Honma, Saitama Cancer Research Institute, 818 Komuro, Ina-machi, Kita-adachi, Saitama 362-0806 Japan
AUTHOR EMAIL: honma@cancer-c.pref.saitama.jp
Cell Growth and Differentiation (CELL GROWTH DIFFER.) (United States) 1999, 10/10 (705-712)
CODEN: CGDIE ISSN: 1044-9523
DOCUMENT TYPE: Journal; Article
LANGUAGE: ENGLISH SUMMARY LANGUAGE: ENGLISH
NUMBER OF REFERENCES: 38

Transforming growth factor beta (TGF-beta) enhanced the growth-inhibitory activities of dexamethasone (Dex) and 1alpha,25-dihydroxyvitamin Dinf 3 (VD3) on human monocytoid leukemia U937 cells. TGF-beta and VD3 synergistically increased the expression of differentiation-associated markers such as the CD11b and CD14 antigens, whereas TGF-beta and Dex did not. On the other hand, TGF-beta and Dex synergistically increased the number of Apo2.7-positive cells, which represents the early stage of apoptosis, whereas TGF-beta and VD3 did not, suggesting that TGF-beta enhanced apoptosis with Dex and enhanced monocytic differentiation with

VD3. In the presence of TGF-beta, the retinoblastoma susceptibility gene product, pRb, was synergistically dephosphorylated by Dex as well as VD3. TGF similarly enhanced the expression of the p21(Waf1) gene in U937 cells **treated** with Dex and VD3. TGF-beta dose- dependently increased the expression of **Bcl-2** and Bad and decreased the expression of Bcl-X(L) in U937 cells. Dex enhanced the down-regulation of Bcl-X(L) expression in TGF-beta-**treated** cells, whereas VD3 blocked this down-regulation of Bcl-X(L). However, the down-regulation of Bcl-X(L) by **treatment** with the **antisense** oligomer did not affect the apoptosis or differentiation of U937 cells. The apoptosis of CD14-positive cells was suppressed in the VD3 plus TGF-beta-**treated** cultures. These results suggest that the expression of CD14 is involved in the survival of differentiated cells.

4/3,AB/127 (Item 3 from file: 72)
DIALOG(R)File 72:EMBASE
(c) 2001 Elsevier Science B.V. All rts. reserv.

07870231 EMBASE No: 1999351230
The endoplasmic reticulum chaperone glycoprotein GRP94 with Casup 2sup +- binding and antiapoptotic properties is a novel proteolytic target of calpain during etoposide-induced apoptosis
Reddy R.K.; Lu J.; Lee A.S.
A.S. Lee, USC/Norris Comprehensive Can. Center, MS 73, Los Angeles, CA 90089 United States
AUTHOR EMAIL: amylee@hsc.usc.edu
Journal of Biological Chemistry (J. BIOL. CHEM.) (United States) 01 OCT 1999, 274/40 (28476-28483)
CODEN: JBCHA ISSN: 0021-9258
DOCUMENT TYPE: Journal; Article
LANGUAGE: ENGLISH SUMMARY LANGUAGE: ENGLISH
NUMBER OF REFERENCES: 52

GRP94 is a 94-kDa chaperone glycoprotein with Casup 2sup +binding properties. We report here that during apoptosis induced by the topoisomerase II inhibitor etoposide, a fraction of GRP94 associated with the endoplasmic reticulum membrane undergoes specific proteolytic cleavage, coinciding with the activation of the caspase CPP32 and initiation of DNA fragmentation. In vivo, inhibitors of caspases able to block etoposide-induced apoptosis can only partially protect GRP94 from proteolytic cleavage, whereas complete inhibition is observed with calpain inhibitor I but not with the proteasome inhibitor. In vitro, GRP94 is not a substrate for CPP32; rather, it can be completely cleaved by calpain, a Casup 2sup +regulated protease. The cleavage of GRP94 by calpain is Casup 2sup +-dependent and generates a discrete polypeptide of 80 kDa. In contrast, calpain has no effect on other stress proteins such as GRP78 or HSP70. Further, immunohistochemical staining reveals specific co-localization of GRP94 with calpain in the perinuclear region following etoposide **treatment**. We further showed that reduction of GRP94 by **antisense** decreased cell viability in etoposide-**treated** Jurkat cells. Our studies provide new evidence that the cytoprotective GRP94, as in the case of the antiapoptotic protein **Bcl-2**, can be targets of proteolytic cleavage themselves during the apoptotic process.

4/3,AB/128 (Item 4 from file: 72)
DIALOG(R)File 72:EMBASE
(c) 2001 Elsevier Science B.V. All rts. reserv.

07857345 EMBASE No: 1999330677
Novel approaches to the **treatment** of small-cell lung cancer
Zangemeister-Wittke U.; Stahel R.A.
U. Zangemeister-Wittke, Division of Oncology, Department of Internal Medicine, University Hospital Zurich, Haldeliweg 4, CH-8044 Zurich

Switzerland
AUTHOR EMAIL: uwe.zan[redacted]eister@dim.usz.ch
Cellular and Molecular Life Sciences (CELL. MOL. LIFE SCI.) (
Switzerland) 1999, 55/12 (1585-1598)
CODEN: CMLSF ISSN: 1420-682X
DOCUMENT TYPE: Journal; Review
LANGUAGE: ENGLISH SUMMARY LANGUAGE: ENGLISH
NUMBER OF REFERENCES: 162

Small-cell lung cancer (SCLC) is characterized by its initial responsiveness to chemotherapy and the appearance of early metastases. Although combination chemotherapy, in some instances together with radiation, has improved the prognosis of this disease, in most patients SCLC ultimately recurs in a drug-resistant form. Several new strategies for the eradication of SCLC are being explored at the preclinical level. The identification of selective target molecules on the surface of SCLC cells, together with the progress made in antibody engineering, have provided new generations of antibodies and immunoconjugates as well as growth factor antagonists and inhibitors. In addition, recent advances in understanding the biology of SCLC have stimulated new investigations searching to counter the molecular basis underlying the increased proliferation and the apoptosis deficiency of SCLC cells. This can be achieved using **antisense** oligodeoxynucleotides that repress the expression of growth factor receptors and anti-apoptosis genes, or by gene replacement to compensate for the loss or inactivation of tumor suppressor genes.

4/3,AB/129 (Item 5 from file: 72)
DIALOG(R)File 72:EMBASE
(c) 2001 Elsevier Science B.V. All rts. reserv.

07853461 EMBASE No: 1999326331
Establishment of an apoptotic model of BGC - 823 gastric cancer cell line with expression of **bcl-2** gene
Ping L.; Junqing C.; Shubao W.
L. Ping, First Hosp. of China Medical Univ., Shenyang China
Chinese Journal of Clinical Oncology (CHIN. J. CLIN. ONCOL.) (China) 1999, 26/8 (586-588)
CODEN: ZZLIE ISSN: 1000-8179
DOCUMENT TYPE: Journal; Article
LANGUAGE: CHINESE SUMMARY LANGUAGE: ENGLISH; CHINESE

Using phase and electronic microscopy, flow cytometry, electrophoresis of DNA the apoptosis of BGC - 823 gastric cancer cell line was observed. The results showed that twenty - four hrs after being **treated** with **bcl - 2 antisense** oligodeoxynucleotides (30uM), Cisplatin (2ug/ml), Carboplantin (20ug/ml), Adriamycin (4ug/ml), and Mitomycin(4ug/ml), the tumor cells shrinked with condensed chromatin, and apoptotic bodies. Hypodiploid DNA was observed by flow cytometry and so did fragmentation of chromosomal DNA by electrophoresis of DNA.

4/3,AB/130 (Item 6 from file: 72)
DIALOG(R)File 72:EMBASE
(c) 2001 Elsevier Science B.V. All rts. reserv.

07853376 EMBASE No: 1999325994
The potential application of **ribozymes** for the **treatment** of hematological disorders
James H.A.
H.A. James, School of Biological Sciences, University of East Anglia, Norwich, Norfolk NR4 7TJ United Kingdom
AUTHOR EMAIL: h.a.james@uea.ac.uk
Journal of Leukocyte Biology (J. LEUKOCYTE BIOL.) (United States) 1999, 66/3 (361-368)

CODEN: JLBIE ISSN: 0741-5400
DOCUMENT TYPE: Journal; Review
LANGUAGE: ENGLISH SUMMARY LANGUAGE: ENGLISH
NUMBER OF REFERENCES: 125

With the identification and increasing understanding of the genes involved in neoplastic transformation has come the realization that abrogation of these genes' products may lead to cell death or a return to normalcy. The use of **ribozymes** and their nucleic acid cousins, **antisense** oligodeoxynucleotides (ODNs), are two such ways of perturbing the disease- related gene expression. This review will look at the development and application of **ribozymes** to abrogate gene expression, with particular relevance to hematological settings. Some examples of **antisense** ODNs will also be mentioned where appropriate.

4/3,AB/131 (Item 7 from file: 72)
DIALOG(R)File 72:EMBASE
(c) 2001 Elsevier Science B.V. All rts. reserv.

07845364 EMBASE No: 1999095317
Antisense bcl-2 treatment increases programmed cell death in non-small cell lung cancer cell lines
Koty P.P.; Zhang H.; Levitt M.L.
M.L. Levitt, Lung Cancer Program, Allegheny University Health Sciences, 320 E. North Avenue, Pittsburgh, PA 15212 United States
Lung Cancer (LUNG CANCER) (Ireland) 1999, 23/2 (115-127)
CODEN: LUCAE ISSN: 0169-5002
PUBLISHER ITEM IDENTIFIER: S016950029800097X
DOCUMENT TYPE: Journal; Article
LANGUAGE: ENGLISH SUMMARY LANGUAGE: ENGLISH
NUMBER OF REFERENCES: 47

Programmed cell death (PCD) is a genetically regulated pathway that is altered in many cancers. This process is, in part, regulated by the ratio of PCD inducers (Bax) or inhibitors (**Bcl-2**). An abnormally high ratio of **Bcl-2** to Bax prevents PCD, thus contributing to resistance to chemotherapeutic agents, many of which are capable of inducing PCD. Non-small cell lung cancer (NSCLC) cells demonstrate resistance to these PCD-inducing agents. If **Bcl-2** prevents NSCLC cells from entering the PCD pathway, then reducing the amount of endogenous **Bcl-2** product may allow these cells to spontaneously enter the PCD pathway. Our purpose was to determine the effects of **bcl-2 antisense treatment** on the levels of programmed cell death in NSCLC cells. First, we determined whether **bcl-2** and bax mRNA were expressed in three morphologically distinct NSCLC cell lines: NCI-H226 (squamous), NCI-H358 (adenocarcinoma), and NCI-H596 (adenosquamous). Cells were then exposed to synthetic **antisense bcl-2** oligonucleotide **treatment**, after which programmed cell death was determined, as evidenced by DNA fragmentation. **Bcl-2** protein expression was detected immunohistochemically. All three NSCLC cell lines expressed both **bcl-2** and bax mRNA and had functional PCD pathways. Synthetic **antisense bcl-2** oligonucleotide **treatment** resulted in decreased **Bcl-2** levels, reduced cell proliferation, decreased cell viability, and increased levels of spontaneous PCD. This represents the first evidence that decreasing **Bcl-2** in three morphologically distinct NSCLC cell lines allows the cells to spontaneously enter a PCD pathway. It also indicates the potential therapeutic use of **antisense bcl-2** in the **treatment** of NSCLC.
Copyright (C) 1999 Elsevier Science Ireland Ltd.

4/3,AB/132 (Item 8 from file: 72)
DIALOG(R)File 72:EMBASE

(c) 2001 Elsevier Science B.V. All rts. reserv.

07830491 EMBASE No: 1999312777
Preclinical and clinical experience of **antisense** therapy for solid tumors
Webb M.S.; Zon G.
M.S. Webb, Inex Pharmaceuticals Corporation, 100-8900 Glenlyon Parkway, Burnaby, BC V5J 5J8 Canada
AUTHOR EMAIL: mwebb@inexpharm.com
Current Opinion in Molecular Therapeutics (CURR. OPIN. MOL. THER.) (United Kingdom) 1999, 1/4 (458-463)
CODEN: CUOTF ISSN: 1464-8431
DOCUMENT TYPE: Journal; Review
LANGUAGE: ENGLISH SUMMARY LANGUAGE: ENGLISH
NUMBER OF REFERENCES: 45

The mechanisms by which the therapeutic window and clinical utility of **antisense** drugs can be fully optimized are discussed. Recent preclinical and clinical efforts are focusing on defining and optimizing the combination therapy regimes in which ONs are most efficacious. However, additional research is required to define which, and how, oncogenes interact with each other and the circumstances under which synergistic therapeutic benefit might be achieved using **antisense** drugs. The therapeutic window of **antisense** drugs is also being expanded by the use of novel delivery systems, including lipid- based carriers for systemic delivery. Taken together, molecular therapeutics based on **antisense** technology, coupled with effective delivery systems increasing drug potency, are anticipated to substantially improve the **treatment** of human neoplasms.

4/3,AB/133 (Item 9 from file: 72)
DIALOG(R)File 72:EMBASE
(c) 2001 Elsevier Science B.V. All rts. reserv.

07750346 EMBASE No: 1999232809
Technology evaluation: G-3139
Banerjee D.
D. Banerjee, Memorial Sloan Kettering Can. Center, 1275 York Avenue, New York, NY 10021 United States
AUTHOR EMAIL: banerjed@aol.com
Current Opinion in Molecular Therapeutics (CURR. OPIN. MOL. THER.) (United Kingdom) 1999, 1/3 (404-408)
CODEN: CUOTF ISSN: 1464-8431
DOCUMENT TYPE: Journal; Review
LANGUAGE: ENGLISH SUMMARY LANGUAGE: ENGLISH
NUMBER OF REFERENCES: 23

G-3139 is an **antisense** phosphorothioate oligodeoxynucleotide (AS PS ON) which suppresses **bcl-2** expression and is being developed by Genta Inc for the potential **treatment** of various cancers [308375]. G-3139 is in various stages of phase I/IIa trials. One study, initiated in May 1999, at the Lombardi Cancer Center at Georgetown University Medical Center, US, will examine G- 3139 in conjunction with docetaxel. In a phase I/IIa dose-escalating trial to **treat** non-Hodgkin's lymphoma (NHL), at the Royal Marsden NHS Trust, UK, no serious, clearly drug-attributable or dose-limiting adverse effects were noted and in some patients encouraging signs of potential drug activity were observed. The responses included one patient in whom cancer mass was reduced and one who developed a complete response for over 38 weeks in duration [239159,291608,325262]. A new phase II protocol using G-3139 combined with standard chemotherapies in relapsed NHL patients has also begun [325262]. Other phase I/IIa studies include: the safety and efficacy of G-3139 in the **treatment** of hormone-resistant, metastatic prostate cancer, when administered with mitoxantrone [305822]; the **treatment** of relapsed follicular NHL, when

administered with cyclophosphamide [311217]; the **treatment** of Stage III and IV metastatic malignant melanoma in combination with dacarbazine [289755]; the **treatment** of hormone-resistant, metastatic prostate cancer when administered over a significantly longer duration than studied previously and in combination with an androgen-receptor blocking agent [291608]. The National Cancer Institute (NCI) funded and conducted preclinical studies of G-3139 in July 1996 and in June 1998, the NCI and Genta entered into a Cooperative Research and Development Agreement (CRADA) for the development of G3139 [290153]. Clinical trials, focusing on colorectal cancer, small cell lung cancer and leukemia, were underway as of April 1999. The company licensed the rights for the use of **bcl-2** as a target for **antisense** and gene therapy-based **treatments** from the University of Pennsylvania. In June 1998, Genta received two patents relating to its **antisense** compounds [289685].

4/3,AB/134 (Item 10 from file: 72)
DIALOG(R)File 72:EMBASE
(c) 2001 Elsevier Science B.V. All rts. reserv.

07674505 EMBASE No: 1999145184
The role of antiapoptotic **Bcl-2** family members in endothelial apoptosis elucidated with **antisense** oligonucleotides
Ackermann E.J.; Taylor J.K.; Narayana R.; Bennett C.F.
E.J. Ackermann, Department of Molecular Pharmacology, Isis Pharmaceuticals, Carlsbad, CA 92008 United States
Journal of Biological Chemistry (J. BIOL. CHEM.) (United States) 16 APR 1999, 274/16 (11245-11252)
CODEN: JBCHA ISSN: 0021-9258
DOCUMENT TYPE: Journal; Article
LANGUAGE: ENGLISH SUMMARY LANGUAGE: ENGLISH
NUMBER OF REFERENCES: 40

In this study, we utilized potent **antisense** oligonucleotides to examine the role of two **Bcl-2** family members found in human umbilical vein endothelial cells (HUVEC). The first, A1, is thought to be a TNF-alpha-inducible cytoprotective gene, and the second, Bcl-XL, is constitutively expressed. Inhibition of the constitutive levels of Bcl-XL caused 10-25% of the cell population to undergo apoptosis and increased the susceptibility of cells to **treatment** with low concentrations of staurosporin or ceramide. The caspase inhibitor benzyloxycarbonyl-Val-Ala-Asp(OMe)CHinf 2 prevented DNA fragmentation and DeltaYm loss caused by Bcl-XL inhibition or Bcl-XL inhibition combined with staurosporin. However, disruption of DeltaYm caused by Bcl-XL inhibition combined with ceramide **treatment** was not inhibited by benzyloxycarbonyl-Val- Ala-Asp(OMe)-CHinf 2, although DNA fragmentation was completely prevented. Taken together, these results demonstrate a direct protective role for Bcl-XL under normal resting conditions and under low level apoptotic challenges to HUVEC. Furthermore, Bcl-XL protects cells from caspase-dependent and -independent mechanisms of DeltaYm disruption. In contrast to Bcl-XL, A1 inhibition did not show a marked effect on the susceptibility of HUVEC to undergo apoptosis in response to TNF-alpha, ceramide, or staurosporin. These results demonstrate that although A1 may be a cytoprotective gene induced by TNF-alpha, it is not primarily responsible for HUVEC resistance to this cytokine.

4/3,AB/135 (Item 11 from file: 72)
DIALOG(R)File 72:EMBASE
(c) 2001 Elsevier Science B.V. All rts. reserv.

07629459 EMBASE No: 1999096136
The endoplasmic reticulum stress-responsive protein GRP78 protects neurons against excitotoxicity and apoptosis: Suppression of oxidative stress and stabilization of calcium homeostasis

Yu Z.; Luo H.; Fu W.; Mattson M.P.
Z. Yu, Sanders-Brown Res. Center on Aging, Dept. of Anatomy and Neurobiology, University of Kentucky, Lexington, KY 40536 United States
Experimental Neurology (EXP. NEUROL.) (United States) 1999, 155/2 (302-314)
CODEN: EXNEA ISSN: 0014-4886
DOCUMENT TYPE: Journal; Article
LANGUAGE: ENGLISH SUMMARY LANGUAGE: ENGLISH
NUMBER OF REFERENCES: 73

The 78-kDa glucose-regulated protein (GRP78) is localized in the endoplasmic reticulum (ER), and its expression is increased by environmental stressors in many types of nonneuronal cells. We report that levels of GRP78 are increased in cultured rat hippocampal neurons exposed to glutamate and oxidative insults (Fe(2+) and amyloid beta-peptide) and that **treatment** of cultures with a GRP78 **antisense** oligodeoxynucleotide increases neuronal death following exposure to each insult. GRP78 **antisense treatment** enhanced apoptosis of differentiated PC 12 cells following NGF withdrawal or exposure to staurosporine. Pretreatment of hippocampal cells with 2-deoxy-D-glucose, a potent inducer of GRP78 expression, protected neurons against excitotoxic and oxidative injury. GRP78 expression may function to suppress oxidative stress and stabilize calcium homeostasis because **treatment** with GRP78 **antisense** resulted in increased levels of reactive oxygen species and intracellular calcium following exposure to glutamate and oxidative insults in hippocampal neurons. Dantrolene (a blocker of ER calcium release), uric acid (an antioxidant), and zVAD-fmk (a caspase inhibitor) each protected neurons against the death-enhancing action of GRP78 **antisense**. The data suggest that ER stress plays a role in neuronal cell death induced by an array of insults and that GRP78 serves a neuroprotective function.

4/3,AB/136 (Item 12 from file: 72)
DIALOG(R)File 72:EMBASE
(c) 2001 Elsevier Science B.V. All rts. reserv.

07474241 EMBASE No: 1998405577
Modulation of apoptosis by endogenous Bcl-x(L) expression in MKN-45 human gastric cancer cells
Kondo S.; Shinomura Y.; Kanayama S.; Higashimoto Y.; Kiyohara T.; Zushi S.; Kitamura S.; Ueyama H.; Matsuzawa Y.
S. Kondo, Second Department Internal Medicine, Osaka University Medical School, 2-2 Yamadaoka, Suita 565-0871 Japan
Oncogene (ONCOGENE) (United Kingdom) 19 NOV 1998, 17/20 (2585-2591)
CODEN: ONCNE ISSN: 0950-9232
DOCUMENT TYPE: Journal; Article
LANGUAGE: ENGLISH SUMMARY LANGUAGE: ENGLISH
NUMBER OF REFERENCES: 32

This study was designed to clarify the role of endogenous Bcl-x(L) expression in modulating apoptosis of malignant cells. Administration of bcl-x-**antisense** oligonucleotides decreased Bcl-x(L) protein levels in the MKN-45 human gastric cancer cell line. The decrease in Bcl-x(L) protein content resulted in increased cell death induced by serum deprivation or Fas-antibody administration. Flow cytometric analysis revealed that the increased apoptotic cell death was more prominent in bcl-x-**antisense**-**treated** cells as compared to control cells, bcl-x-sense-**treated** cells, or bcl-x-nonsense-**treated** cells. To inhibit the effect of intrinsic Bcl-x(L) protein, we overexpressed Bak, which binds Bcl-x(L), and inhibits the anti-apoptotic effect of Bcl-x(L), by transfection into MKN-45 cells. Bak-overexpressing cells showed increased apoptotic cell death induced by Fas-antibody when compared to parent cells and MKN-neo-transfected cells. Bak-overexpressing cells also showed greater sensitization to 5-fluorouracil and cisplatin than parent cells and MKN-neo-transfected cells. In conclusion, we demonstrated that

administration of bcl-x **antisense** oligonucleotides or overexpression of Bak protein induces [redacted] sitization to apoptosis in MKN[redacted] gastric cancer cells, suggesting that endogenous Bcl-x(L) expression in cancer cells is an important modulator of apoptosis.

4/3,AB/137 (Item 13 from file: 72)
DIALOG(R)File 72:EMBASE
(c) 2001 Elsevier Science B.V. All rts. reserv.

07449928 EMBASE No: 1998366411
Vascular gene transfer for the **treatment** of restenosis and atherosclerosis
Laitinen M.; Yla-Herttuala S.
Dr. S. Yla-Herttuala, A.I. Virtanen Institute, University of Kuopio, PO Box 1627, FIN-70211 Kuopio Finland
AUTHOR EMAIL: Ylahertt@keula.uku.fi
Current Opinion in Lipidology (CURR. OPIN. LIPIDOLOGY) (United Kingdom) 1998, 9/5 (465-469)
CODEN: COPLE ISSN: 0957-9672
DOCUMENT TYPE: Journal; Review
LANGUAGE: ENGLISH SUMMARY LANGUAGE: ENGLISH
NUMBER OF REFERENCES: 61

Local gene transfer into the vascular wall offers a promising alternative to **treat** atherosclerosis-related diseases at cellular and molecular levels. Blood vessels are among the easiest targets for gene therapy because of novel percutaneous, catheter-based **treatment** methods. On the other hand, gene transfer to the artery wall can also be accomplished from adventitia, and in some situations intramuscular gene delivery is also a possibility. In most conditions, such as postangioplasty restenosis, only a temporary expression of the transfected gene will be required. Promising therapeutic effects have been obtained in animal models of restenosis with the transfer of genes for vascular endothelial growth factor, fibroblast growth factor, thymidine kinase, p53, bcl-x, nitric oxide synthase and retinoblastoma. Also, growth arrest homeobox gene and **antisense** oligonucleotides against transcription factors or cell cycle regulatory proteins have produced beneficial therapeutic effects. Angiogenesis is an emerging new target for gene therapy of ischemic diseases. In addition, hyperlipoproteinemias may be improved by transferring functional lipoprotein-receptor genes into hepatocytes of affected individuals. First experiences of gene transfer methods in the human vascular system have been reported. However, further studies regarding gene delivery methods, vectors and safety of the procedures are needed before a full therapeutic potential of gene therapy in vascular diseases can be evaluated.

4/3,AB/138 (Item 14 from file: 72)
DIALOG(R)File 72:EMBASE
(c) 2001 Elsevier Science B.V. All rts. reserv.

07409000 EMBASE No: 1998287654
Antisense comes of age
Flanagan W.M.
W.M. Flanagan, 333 Lakeside Drive, Foster City, CA 94404 United States
Cancer and Metastasis Reviews (CANCER METASTASIS REV.) (Netherlands) 1998, 17/2 (169-176)
CODEN: CMRED ISSN: 0167-7659
DOCUMENT TYPE: Journal; Review
LANGUAGE: ENGLISH SUMMARY LANGUAGE: ENGLISH
NUMBER OF REFERENCES: 45

During the last ten years, **antisense** technology has experienced growing pains not unlike those of adolescence. In 1992, **antisense** was trumpeted as one of the top 10 emerging research areas. However, 3 years

later, researchers were confronted with significant problems associated with **antisense** oligonucleotides ranging from sequence-dependent, non-**antisense** effects in vitro to dose-limiting toxicities in preclinical models [1-3]. Many researchers had doubts whether sequence-specific **antisense** even existed or whether it would ever exist as a therapeutic strategy [4]. Despite these gloomy predictions, many of the challenges facing the development of **antisense**-based drugs as therapeutics have been overcome as evidenced by the progress of several **antisense** oligonucleotides in the clinic for the **treatment** of cancer.

4/3,AB/139 (Item 15 from file: 72)
DIALOG(R)File 72:EMBASE
(c) 2001 Elsevier Science B.V. All rts. reserv.

07380760 EMBASE No: 1998289166
Interference of apoptosis with the **treatment** of malignant blood diseases: The example of BCL2 proteins
INTERFERENCE DES ANOMALIES DE L'APOPTOSE AVEC LES TRAITEMENTS DES HEMOPATHIES MALIGNES: L'EXEMPLE DES PROTEINES **BCL 2**
Larsen C.-J.
C.-J. Larsen, CNRS - IBMIG, 40 avenue du Recteur Pineau, 86020 Poitiers France
Hematologie (HEMATOLOGIE) (France) 1998, 4/3 SUPPL. 2 (76-79)
CODEN: HEMAF ISSN: 1264-7527
DOCUMENT TYPE: Journal; Short Survey
LANGUAGE: FRENCH
NUMBER OF REFERENCES: 17

4/3,AB/140 (Item 16 from file: 72)
DIALOG(R)File 72:EMBASE
(c) 2001 Elsevier Science B.V. All rts. reserv.

07206745 EMBASE No: 1998077137
Molecular abnormalities in lung cancer
Salgia R.; Skarin A.T.
Dr. R. Salgia, Dana-Farber Cancer Institute, Division of Adult Oncology, Dana 530C, 44 Binney St, Boston, MA 02115 United States
Journal of Clinical Oncology (J. CLIN. ONCOL.) (United States) 1998, 16/3 (1207-1217)
CODEN: JCOND ISSN: 0732-183X
DOCUMENT TYPE: Journal; Review
LANGUAGE: ENGLISH SUMMARY LANGUAGE: ENGLISH
NUMBER OF REFERENCES: 148

Purpose: To review several recently described molecular abnormalities in lung cancer and discuss their potential diagnostic and therapeutic relevance. Design: Articles were identified through a Medline search (1966 to 1997) and studies, including reviews, were cited in the references. Results: Molecular mechanisms altered in lung cancer include induced expression of oncogenes, such as RAS, MYC, c-erbB-2, and **BCL-2**, and loss of tumor-suppressor genes, such as RB, p53, and p 16(INK4A). RAS is a 21-kd G protein and up to 30% of adenocarcinomas show mutations in K-RAS oncogene. MYC encodes a transcriptional activator and amplification may adversely affect survival in small-cell lung cancer (SCLC). The growth factor receptor c-erbB-2 is overexpressed in up to 25% of non-small-cell lung cancer (NSCLC) cases. **BCL- 2**, a negative regulator of apoptosis, is expressed differently in some NSCLCs. Abnormalities of RB, a key regulator of cell cycle, are detected in greater than 90% of SCLCs. There is an inverse relationship in lung cancer cells between expression of RB and p16(INK4A), an upstream regulator of RB. Mutations of p53, with frequencies up to 50% in NSCLC and 80% in SCLC, can lead to loss of tumor-suppressor function, cellular proliferation, and inhibition of

apoptosis. The identified molecular abnormalities in lung cancer are currently used to develop diagnostics for detecting early disease, as well as to identify targets for gene therapy. Conclusion: Genetic abnormalities involved in the pathogenesis of lung cancer are rapidly being delineated. Understanding molecular abnormalities in lung cancer could potentially lead to earlier diagnosis and the development of novel investigational approaches to the **treatment** of lung cancer.

4/3,AB/141 (Item 17 from file: 72)
DIALOG(R)File 72:EMBASE
(c) 2001 Elsevier Science B.V. All rts. reserv.

07199160 EMBASE No: 1998096139
Oncogenes and tumor suppressor genes: Therapeutic implications
Stass S.A.; Mixson A.J.
S.A. Stass, University of Maryland, Department of Pathology, Greenbaum Cancer Center, 22 South Greene Street, Baltimore, MD 21201 United States
Clinical Cancer Research (CLIN. CANC. RES.) (United States) 1997, 3/12 II (2687-2695)
CODEN: CCREF ISSN: 1078-0432
DOCUMENT TYPE: Journal; Conference Paper
LANGUAGE: ENGLISH SUMMARY LANGUAGE: ENGLISH
NUMBER OF REFERENCES: 58

Genetic instability is a hallmark of cancer. Alterations in DNA through mutations, deletions, and translocations affect genes that are fundamental to normal tell growth differentiation and programmed cell death. Here, we discuss these alterations as they relate to oncogenes and tumor suppressor genes. In addition, we describe the implications the changes in oncogenes and tumor suppressor genes have on designing new therapeutic strategies for the **treatment** of cancer.

4/3,AB/142 (Item 18 from file: 72)
DIALOG(R)File 72:EMBASE
(c) 2001 Elsevier Science B.V. All rts. reserv.

07179809 EMBASE No: 1998069824
1,25-dihydroxyvitamin Dinf 3 protects human leukemic cells from tumor necrosis factor-induced apoptosis via inactivation of cytosolic phospholipase Ainf 2
Wu Y.-L.; Jiang X.-R.; Lillington D.M.; Allen P.D.; Newland A.C.; Kelsey S.M.
S.M. Kelsey, Dept. of Hematology, St. Bartholomew's/Royal London, School of Medicine and Dentistry, Turner St., London E1 2AD United Kingdom
AUTHOR EMAIL: S.M.Kelsey@mds.qmw.ac.UK
Cancer Research (CANCER RES.) (United States) 15 FEB 1998, 58/4 (633-640)
CODEN: CNREA ISSN: 0008-5472
DOCUMENT TYPE: Journal; Article
LANGUAGE: ENGLISH SUMMARY LANGUAGE: ENGLISH
NUMBER OF REFERENCES: 47

The mechanism by which tumor necrosis factor (TNF) induces death of cancer cells appears to involve the activation of cytosolic phospholipase Ainf 2 (cPLAinf 2). U937 human leukemic cells **treated** with 1,25-dihydroxyvitamin Dinf 3 (1,25(OH)inf 2Dinf 3; 10sup -sup 8 M) become resistant to TNF, an effect that is independent of cell cycle status and expression of TNF receptors or **BCL-2**. In this study, TNF produced a dose- and time-dependent enhancement of (sup 3H)arachidonic acid release in U937 cells. The amount of (sup 3H)arachidonic acid release was positively associated with TNF-induced apoptosis. Both immunofluorescence microscopy and Western blotting of cell subcompartments demonstrated translocation of cPLAinf 2 from the cytosol to the cell membrane in

response to TNF. In addition, TNF up-regulated expression of cPLAinf 2 mRNA. An **antisense** oligonucleotide to cPLAinf 2 and the cPLAinf 2 inhibitor 4-bromophenacyl bromide significantly inhibited TNF-induced cytotoxicity. Prior incubation of cells with 1,25(OH)inf 2Dinf 3 significantly inhibited (a) TNF-induced (sup 3H)arachidonic acid release and apoptosis, (b) TNF-induced translocation of cPLAinf 2 to the membrane, and (c) the up-regulation of cPLAinf 2 mRNA with TINT. Furthermore, the inhibitory effect of 1,25(OH)inf 2Dinf 3 was not reversed by inhibitors of transcription or translation. The data suggest that activation of cPLAinf 2 is involved in TNF-induced apoptosis of leukemic cells. 1,25(OH)inf 2Dinf 3 directly inhibits cPLAinf 2 translocation and mRNA up-regulation induced by TNF. Disruption of cPLAinf 2 activation may represent a possible mechanism whereby leukemic cells can become resistant to TNF-mediated killing.

4/3,AB/143 (Item 1 from file: 159)
DIALOG(R)File 159:Cancerlit
(c) format only 2001 Dialog Corporation. All rts. reserv.

01504847 99196917
Induction of p21(CIP1/Waf1) and activation of p34(cdc2) involved in retinoic acid-induced apoptosis in human hepatoma Hep3B cells.
Hsu SL; Chen MC; Chou YH; Hwang GY; Yin SC
Department of Education & Research, Taichung Veterans General Hospital, Taichung, 40705, Taiwan. h2326@vghtc.vghtc.gov.tw
Exp Cell Res; 248(1):87-96 1999 ISSN 0014-4827 Journal Code: EPB
Languages: ENGLISH
Document Type: JOURNAL ARTICLE
The biological activity of retinoic acid (RA) was examined in human hepatoma Hep3B cells. Under serum-deprived conditions, RA induced S/M-phase elevation and mitotic index increase within 24 h, followed by apoptosis. This RA-induced apoptosis was accompanied by p53-independent up-regulation of endogenous p21(CIPI/Waf1) and Bax proteins, as well as activation of p34(cdc2) kinase, and increase of Rb2 protein level and phosphorylation pattern. In addition, RA had no effect on the levels of Bcl-XL; Bcl-XS; cyclins A, B, D1, D3, or E; or Rb1 expression but markedly down-modulated Cdk2 kinase activity and reduced Cdk4 expression. RA also slightly delayed p27(Kip1) expression. Olomoucine, a potent p34(cdc2) and Cdk2 inhibitor, effectively blocked RA-mediated p34(cdc2) kinase activation and prevented RA-induced apoptosis. Furthermore, **antisense** oligonucleotide complementary to p21(CIP2/Waf1) and p34(cdc2) mRNA significantly rescued RA-induced apoptosis. Our data indicate that p21(CIP2/Waf1) overexpression may not be the only regulatory factor necessary for RA-induced apoptosis in human hepatoma Hep3B cells. RA **treatment** leads to Rb2 hyperphosphorylation, and p34(cdc2) kinase activation is coincident with an aberrant mitotic progression, followed by appearance of abnormal nucleus. This aberrant cell cycle progression appeared requisite for RA-induced cell death. These findings suggest that inappropriate regulation of the cell cycle regulators p21(CIP2/Waf1) and p34(cdc2) is coupled with induction of Bax and involved in cell death with apoptosis when Hep3B cells are exposed to RA. Copyright 1999 Academic Press.

4/3,AB/144 (Item 2 from file: 159)
DIALOG(R)File 159:Cancerlit
(c) format only 2001 Dialog Corporation. All rts. reserv.

01490528 99702042
A Phase I-II Study with Dacarbazine and **BCL-2 Antisense** Oligonucleotide G3139 (GENTA) as a Chemosensitizer in Patients with Advanced Malignant Melanoma (Meeting abstract).
Jansen Burkhar; Wacheck Volke; Heere-Ress Elisabet; Schlagbauer-Wadl Hermin; Hollenstein Ursul; Lucas Trevo; Eichler Hans-Georg; Wolff Klau; Pehamberger Huber
Dept. of Clinical Pharmacology / Section of Experimental Oncology

University of Vienna, Vienna, Austria.
Proc Annu Meet Am Soc [redacted]in Oncol; 18:A2049 1999
Languages: ENGLISH
Document Type: MEETING ABSTRACTS
Chemoresistance of malignant melanoma has been linked to an upregulation of **BCL-2** expression which is found in up to 90% of all cases. Recently we showed in a SCID mouse melanoma model, that **antisense** oligonucleotide G3139 (Genta Inc.) directed against **BCL-2** enhanced tumor cell apoptotis and decreased growth of human melanoma xenotransplants; cotreatment with dacarbazine (DTIC) caused dramatic enhancement of antitumor effects (Jansen et al., Nature Medicine 4:232, 1998). In the present Phase I-II study we are evaluating combined therapy of dacarbazine and G3139 in patients with advanced malignant melanoma, including patients with disease resistant to prior single-agent dacarbazine. G3139 is adminstered as a two week continuous intravenous infusion in addition to a standard dacarbazine regimen (200 mg/m2/X 5 days). 3-patient cohorts are **treated** with escalating doses of 0.6, 1.3, 1.7, and 2.3 mg/kg day for 14 days, followed by a 2-week rest. To date, intravenous infusion of G3139 and dose escalation is well tolerated up to and including 2.3 mg/kg/day without dose limiting adverse events; dose escalation is continuing. Serial measurements of **BCL-2** protein levels in melanoma biopsies have demonstrated reduction in **BCL-2** expression coincident with G3139 therapy. Disappearance of select metastatic lesions has been observed. To date, no patients have discontinued G3139 therapy due to adverse events. Our initial results of this Phase I-II study indicate that G3139 can reduce **BCL-2** expression in metastatic melanoma, and combined therapy with dacarbazine is well tolerated. Clinical therapy with G3139 **antisense** oligonucleotide targeting **BCL-2** expression, may be a novel and rational approach to improve response to chemotherapy in this or other malignancies where chemoresistance is linked to high **BCL-2** levels. (C) American Society of Clinical Oncology 1999.

4/3,AB/145 (Item 3 from file: 159)
DIALOG(R)File 159:Cancerlit
(c) format only 2001 Dialog Corporation. All rts. reserv.

01489725 99701239
A Phase I/IIA Dose-Escalating Trial of **bcl-2 Antisense** (G3139) **Treatment** by 14-Day Continuous Intravenous Infusion (CI) for Patients with Androgen-Independent Prostate Cancer or Other Advanced Solid Tumor Malignancies. (Meeting abstract).
Morris Michael; Tong Willia; Osman Ima; Maslak Pete; Kelly William; Terry Kathry; Rosen Nea; Scher Howard
Memorial Sloan-Kettering Cancer Center, New York, NY.
Proc Annu Meet Am Soc Clin Oncol; 18:A1243 1999
Languages: ENGLISH
Document Type: MEETING ABSTRACTS
The **bcl-2** protooncogene encodes an inner mitochondrial protein that inhibits apoptosis. **Bcl-2** overexpression has been implicated in the growth and chemoresistance of a variety of solid tumors including breast, lung, kidney, and androgen-independent prostate cancers. G3139 (Genta Incorporated) is an 18-base **bcl-2 antisense** oligonucleotide shown to induce regression of relapsed lymphomas when given as a 14-day subcutaneous infusion. Preclinical data indicate that therapeutic effects are seen when plasma concentrations are approximately 1 ug/ml or greater. In this trial, patients with advanced solid tumors received escalating doses of G3139 by continuous intravenous infusion with an objective of determining the optimal biologic dose based on changes in **bcl-2** expression in peripheral blood lymphocytes (PBL's). 15 evaluable patients (11 with prostate, 2 with renal cell, and 2 with prostate and renal cell cancers) received G3139 for 14 days followed by 4 weeks of observation. Cohorts of three patients each received 0.6, 1.3, 1.7 mg/kg/d; 6 patients received 2.3 mg/kg/d. No toxicities exceeded grade 1-2,

except one patient at the 2.3 mg/kg/day level who developed grade 3 neutropenia that resolved in 24 hrs; enrollment continued at higher levels. Two patients with renal cell and one with prostate cancer showed no progression with 3, 3 and 2 **treatments** respectively. Mean steady-state plasma levels were <1 ug/ml in patients **treated** at 0.6-1.7 mg/kg/d, and 1.2 ug/ml at 2.3 mg/kg/d. These results indicate that G3139 is well tolerated intravenously as monotherapy by CI in patients with advanced solid tumors. Although concentrations that have been shown to inhibit **bcl-2** expression have been achieved, the optimal biologic dose has not been defined measuring apoptotic changes in PBL's. The safety data support further clinical development of G3139, both alone and in combination with chemotherapy. Supported by Genta Corporation, CaPCURE, CA 05826 and CA09207. (C) American Society of Clinical Oncology 1999.

4/3,AB/146 (Item 4 from file: 159)
DIALOG(R)File 159:Cancerlit
(c) format only 2001 Dialog Corporation. All rts. reserv.

01317310 98642054
First demonstration of anti-lymphoma activity of **bcl-2** **antisense** molecule G3139; results of phase I/IIA clinical trial (Meeting abstract).
Anonymous
Royal Marsden Hospital (RMH), UK
Proc Annu Meet Am Soc Clin Oncol; 16:A54 1997 ISSN 0732-183X
Languages: ENGLISH
Document Type: MEETING ABSTRACTS; CLINICAL TRIAL; CLINICAL TRIAL, PHASE I ; CLINICAL TRIAL, PHASE II
It has been well known that T14/18 translocation in follicular lymphoma up-regulates **BCL-2** leading to continued expression of **BCL-2** protein. Upregulation of **BCL-2** leads to extended survival of the cells and increased chemoresistance. Clinical trials demonstrated correlation between **BCL-2** expression and poor clinical prognosis in an intermediate and high grade lymphomas. G3139 is an all-phosphorothioate 18mer oligonucleotides targeted to the first six codons of the **BCL-2** mRNA. It has been shown to specifically down regulate **BCL-2** in vitro and to have dose dependent activity in mice models of human lymphoma as well as other xenograft models of solid tumours. The Lymphoma Unit at the RMH performed the first phase I trial in all grades NHL patients (pts) who relapsed following several previous conventional chemotherapy regimens and who expressed **BCL-2**. Replicating preclinical xenograft model, the pts received G3139 as a continuous subcutaneous 14 day infusion. The doses were escalated according to EORTC scheme and safety as well as efficacy measured using standard evaluation criteria. Until early December 1996, 10 pts were entered in 6 dose escalation cohorts up to a dose of 147.6 mg/m2/day. Based on excellent systemic tolerance and lack of even grade II drug attributable toxicities, the escalations were made in 100% increments. There was mild topical, infusion site irritation which was acceptable in all but one pt. Blood levels of two pts at 5th escalation level approximated concentration effective in in vivo models of lymphoma. Four pts demonstrated improvement in disease status as defined by clinical and/or laboratory parameters including decrease in **BCL-2** protein. One of those 4 pts demonstrated minor tumour response. Another pt on the higher dose, who failed 4 prior therapies, with diagnosis of Working Formulation C, mixed small cleaved and large cell, stage IVB, developed complete clinical and radiological response of 16+ week duration. We conclude that **antisense** approach to **BCL-2** constitutes a potentially important **treatment** modality in NHL, leading to response in poor prognosis pts at doses causing less than Grade II toxicity. The trial is continuing and the full update will be presented. (C) American Society of Clinical Oncology 1997.

4/3,AB/147 (Item 5 from file: 159)
DIALOG(R)File 159:Cancerlit
(c) format only 2001 Dialog Corporation. All rts. reserv.

01317121 98641199
The evidence for endogenous galectin-3 to inhibit cisplatin-induced apoptosis in breast cancer cell (Meeting abstract).
Akahani S; Inohara H; Nangia-Makker P; Silletti SA; Raz A
Tumor Progression and Metastasis, Karmanos Cancer Institute, Wayne State University, Detroit, MI 48201
Proc Annu Meet Am Assoc Cancer Res; 38:A4199 1997 ISSN 0197-016X
Languages: ENGLISH
Document Type: MEETING ABSTRACTS
Galectin-3, a beta-galactoside binding protein with 31 kDa of molecular weight, has been considered that it may play key roles in tumor progression and metastasis. In this study, we investigated the possible function of galectin-3 in cis-platinum(II) diammine dichloride (cisplatin-CDDP) induced apoptosis. As reported previously, transfection of galectin-3 cDNA resulted in the acquisition of tumorigenic activity in non-tumorigenic human breast carcinoma BT549. Here BT549 sense transfectant with galectin-3 displayed the increased resistance to CDDP **treatment** in a time and dose-dependent manner. After continuous exposure to CDDP, galectin-3 free wildtype BT549 and BT549 **antisense** transfectant exhibited chromatin condensation and DNA fragmentation which is characterizing apoptosis in epithelial cells, whereas BT549 sense transfectant did not. In addition, wildtype BT549, sense and **antisense** transfectant showed no expression of **bcl-2**, bcl-x, and bax before and after CDDP exposure. Hence it has been indicated that endogenous galectin-3 may suppress the apoptotic response of tumor cells in a **bcl-2** independent pathway.

4/3,AB/148 (Item 6 from file: 159)
DIALOG(R)File 159:Cancerlit
(c) format only 2001 Dialog Corporation. All rts. reserv.

01314118 98638140
Antisense oligonucleotides targeting sequences shared by the **Bcl-2** and the Bcl-xL mRNA efficiently downregulate expression of both proteins and induce apoptosis of lung cancer cells (Meeting abstract).
Luedke GH; Ziegler A; Stahel RA; Zangemeister-Wittke U
Division of Oncology, Department of Internal Medicine, University Hospital, CH-8091 Zurich, Switzerland
Proc Annu Meet Am Assoc Cancer Res; 38:A1140 1997 ISSN 0197-016X
Languages: ENGLISH
Document Type: MEETING ABSTRACTS
Lung cancer represents a major cause of cancer-related death and its incidence continues to rise worldwide. Whereas non-small cell lung cancer (NSCLC) is inherently resistant to initial chemotherapy, drug resistance of small cell lung cancer (SCLC) is generally acquired during **treatment**. Resistance to apoptosis has emerged as an important category of drug resistance also in lung cancer, and likely explains a significant proportion of **treatment** failures. Examination of SCLC and NSCLC cell lines revealed different basal levels of **Bcl-2** and Bcl-xL protein expression, which was dependent on the histomorphological classification. Whereas SCLC cells expressed detectable levels of **Bcl -2** , but very low levels of Bcl-xL, the opposite constellation was found in NSCLC cells. With the aim to restore the apoptotic response of SCLC and NSCLC cells, a strategy based on the use of 20-mer **antisense** oligodeoxynucleotides (ODNs) targeting different sequences shared by the **Bcl-2** and the Bcl-xL, but not the Bcl-xS mRNA, was examined. As determined by Western blot and cytotoxicity analysis, **treatment** of cells with two ODNs complementary to the coding regions of both mRNA species efficiently downregulated expression of both anti-apoptosis

proteins in a dose-dependent manner, and induced apoptosis in up to 85% of lung cancer cells, independent of their histomorphological phenotype. Scrambled control ODNs did not alter **Bcl-2** and Bcl-xL levels, nor did they reduce cell viability. The data suggest the use of **antisense** ODNs which simultaneously downregulate **Bcl-2** and Bcl-xL expression as a novel **treatment** for all types of lung cancer.

4/3,AB/149 (Item 7 from file: 159)
DIALOG(R)File 159:Cancerlit
(c) format only 2001 Dialog Corporation. All rts. reserv.

01314116 98638138
Bcl-2 selectively inhibits nuclear import of p53 after genotoxic damage (Meeting abstract).
Beham A; Marin M; Herrmann JL; Fernandez A; Lozano G; McDonnell TJ
The University of Texas M.D. Anderson Cancer Center, Houston, TX 77030
The University of Texas M.D. Anderson Cancer Center, Houston, TX 77030
Proc Annu Meet Am Assoc Cancer Res; 38:A1138 1997 ISSN 0197-016X
Languages: ENGLISH
Document Type: MEETING ABSTRACTS
Cell stress signals including irradiation, TNFalpha, and heat shock up-regulate or activate specific proteins including p53 and NFkappaB. These proteins are dependent on nuclear localizing signal (NLS) mediated translocation to function. Radiation induction and nuclear import of p53 protein resulted in cell death in control LNCaP prostate cancer cells as assessed by immunoblotting and confocal microscopy. In contrast, p53 protein was induced but did not undergo nuclear translocation in **bcl-2** congenic cells. This effect was correlated with a significant reduction in the ability of p53 to transactivate a promoter construct possessing a p53 binding site. RKO colon carcinoma cells also possess wt-p53 and high levels of **bcl-2**. A 3-fold reduction in **bcl-2** was observed in RKO cells using liposomal **antisense** **bcl-2** oligonucleotides. This was associated with nuclear import of p53 and correlated with acquisition of radiosensitivity compared to cells **treated** with control oligos. Control, but not **bcl-2** congenic, LNCaP cells were sensitive to cell death induction by TNFalpha. TNFalpha activation, and nuclear import, of NFkappaB in control and **bcl-2** expressing cells was similar. It has been shown that NFkappaB activation by TNFalpha is not required for the mediation of TNFalpha induced cell death. We propose that the selective regulation of NLS mediated transmembrane traffic by **bcl-2** may be important for the inhibition of cell stress induced apoptosis.

4/3,AB/150 (Item 8 from file: 159)
DIALOG(R)File 159:Cancerlit
(c) format only 2001 Dialog Corporation. All rts. reserv.

01231422 97620831
Molecular mechanisms of oncogenesis.
Anonymous
No affiliation given
Non-serial; Molecular Mechanisms of Oncogenesis, 5th Hanson Symposium. Adelaide, Australia, November 4-7, 1996: 1996
Languages: ENGLISH
Document Type: MONOGRAPH
Titles of the sessions in this Symposium were: receptor activation, dealing with cytokines, differentiation signaling, activating mutations in cytokine common beta chains, signal-regulated kinases, the Bcr/Abl oncogene, cKit transformation, cytokine receptor activation, and macrophage differentiation; intracellular signalling, with topics dealing with the EGF receptor, Raf-1 activation, 14-3-3 proteins, pathways involving Src tyrosine kinases, analyses using CSF knockout mice, the Sos1 ras-activating protein and the JAK/STAT pathway; tumor invasion, including talks on

adhesion-mediated signaling, the roles of sulfated oligosaccharides, the cell matrix and cell-cell interactions, p53 gene therapy of cancer, transgenic mouse models of skin carcinogenesis, the chemokine gene family, and betal integrin; apoptosis, covering its nature and implications for oncology, selective proteolysis during apoptosis, DNA damage and fragmentation, regulation of the process in the thymus, and the influence of **Bcl-2**, granzyme B inhibitor, and apoptosis stimulated by the GM-CSF analog E21R; transcription factors as oncogenes; cell cycle regulation; new approaches to cancer **treatment**, specifically peptide agonists of erythropoietin, use of **ribozymes** , manipulation of hemopoietic tissue, and cationic lipid transfection molecules; selected abstracts on cell surface proteins, macrophage inflammatory protein-a, regulation by the interleukin-2 receptor, the common signaling pathway for CD40 and p75 TNF and the scl gene; and tumor genetics.

4/3,AB/151 (Item 9 from file: 159)
DIALOG(R)File 159:Cancerlit
(c) format only 2001 Dialog Corporation. All rts. reserv.

01230877 97610458
NSAID-induced apoptosis in v-src transformed CEFs is dependent on c-myc (Meeting abstract).
Lu X; Fairbairn DW; Bradshaw WS; O'Neill KL; Ewert DL; Simmons DL
Department of Zoology, Brigham Young Univ., Provo, UT 84602
Proc Annu Meet Am Assoc Cancer Res; 37:A4116 1996 ISSN 0197-016X
Languages: ENGLISH
Document Type: MEETING ABSTRACTS
We have recently shown NSAIDS apoptosis in v-src-transformed chicken embryo fibroblasts (CEF). Here we report that for diclofenac, commitment to cell death occurs earlier and at doses lower than previously thought. Although v-src antagonizes apoptosis in most systems, we show that this oncogene alone can induce apoptosis in CEF upon withdrawal of trophic factors from the medium. In contrast, NSAID-induced apoptosis (NIA) is absolutely dependent upon the expression of v-src, but is independent of the presence of trophic factors. Expression of three apoptosis-related genes c-rel,p53 and **bcl-2** remains unchanged after drug **treatment**. NIA in cells in which human **bcl-2** is overexpressed, however, is inhibited as much as 10-fold. In contrast, there is a persistent, greater than 10-fold induction of c-myc mRNA during NIA. Moreover, transfection of **antisense** c-myc oligonucleotides produced a 50% inhibition of apoptosis, suggesting that NIA in v-src transformed CEF proceeds through a c-myc dependent pathway.

4/3,AB/152 (Item 10 from file: 159)
DIALOG(R)File 159:Cancerlit
(c) format only 2001 Dialog Corporation. All rts. reserv.

01228975 97604994
Gene therapy of advanced prostate cancer by in vivo transduction with prostate-targeted **antisense** c-myc RNA retroviruses (Meeting abstract).
Steiner MS; Anthony CT; Lu Y; Smith JA Jr; Moses HL; Holt JT
Dept. of Urology, Univ. of Tennessee, Memphis, TN 38163
Proc Annu Meet Am Assoc Cancer Res; 37:A2349 1996 ISSN 0197-016X
Languages: ENGLISH
Document Type: MEETING ABSTRACTS
A novel strategy to combat advanced prostate cancer utilizes replication in competent retroviruses containing mouse mammary tumor virus (MMTV) tissue-specific promoters to allow selective transcription of therapeutic genes within prostate tumor cells. Androgen refractory prostatic cancer frequently has overexpression of the proto-oncogene c-myc which contributes to uncontrolled prostate cancer proliferation. Consequently, cultured human prostate cancer DU145 cells were transduced by XM6:MMTV-**antisense**

c-myc retroviruses (titer 7 x 10(5) virions/ml). Although DU145 cell proliferation in culture was unchanged, a single direct injection of MMTV-**antisense** c-myc retroviral media into established DU145 tumors in nude mice produced a 94.5% reduction in tumor size compared to tumors **treated** with control virus MMTV lacking **antisense** c-myc and untreated tumor by 70 days (MMTV **antisense** c-myc 13.2 +/- 3.6 mm3, n = 10 versus control 182.3 +/- 38.5 mm3, n = 9 and untreated 238.2 +/- 95 mm3, n = 8; p less than 0.004). Histopathological MMTV-**antisense** c-myc transduced DU145 tumors showed increased tumor cell differentiation, decreased invasion and a marked stromal response. The mechanism for the antitumor effect of MMTV **antisense** c-myc retrovirus appears to be suppression of c-myc mRNA and protein, decreased **bcl-2** protein, and apoptosis. Thus, the in vivo transduction of prostate cancer cells with MMTV-**antisense** c-myc retroviruses reduced tumor growth, and may be useful for gene therapy against advanced prostate cancer.

4/3,AB/153 (Item 11 from file: 159)
DIALOG(R)File 159:Cancerlit
(c) format only 2001 Dialog Corporation. All rts. reserv.

01225893 96653704
Antitumor activity of liposomal-**bcl-2-antisense** oligonucleotides in follicular lymphoma (Meeting abstract).
Tormo M; Tari A; McDonnell TJ; Khodadadian M; Cabanillas F; Garcia-Conde J; Lopez-Berestein G
Univ. Texas MD Anderson Cancer Center, Houston, TX 77030
Proc Annu Meet Am Assoc Cancer Res; 37:A1190 1996 ISSN 0197-016X
Languages: ENGLISH
Document Type: MEETING ABSTRACTS
Approximately 90% of follicular lymphomas (FL) have the t(14;18) translocation, resulting in overexpression of **bcl-2** protein. **Bcl-2** is an oncogene with tumorigenic potential due to its capacity to block apoptosis. P-ethoxy-oligonucleotide, a non-ionic and nuclease-resistant phosphodiester analog, was used as an **antisense** molecule. **Antisense** oligonucleotide complementary to the first open reading frame of the **bcl-2** mRNA and two control oligonucleotides were incorporated into liposomes. We studied the effect of liposomal-**bcl-2 antisense** oligonucleotide (L-**bcl-2**) on the growth of a FL cell line with t(14;18) translocation (Johnson cells) and 2 cell lines without the translocation but with overexpression of **bcl-2** protein (Raji and Jurkat cells). 75% growth inhibition was achieved at 5 uM of L-**bcl-2** in Johnson cells, but not in Raji and Jurkat cells. No activity were observed with empty liposomes. On the other hand, there was a dose dependent decrease in **bcl-2** protein in all cell lines **treated** with L-**bcl-2** as measured by immunoblot analysis. One of the mechanisms by which L-**bcl-2** growth inhibition is mediated in Johnson cells might be through induction of cells into apoptosis, since the **treated** cells have an increased apoptotic index as measured by the acridine orange method. These studies indicate that L-**bcl-2** might form the basis for a molecular approach to follicular lymphoma therapy.

4/3,AB/154 (Item 12 from file: 159)
DIALOG(R)File 159:Cancerlit
(c) format only 2001 Dialog Corporation. All rts. reserv.

01224786 96625720
TNF-induced FGF-1 upregulation prevents apoptosis in transformed endothelial cells (Meeting abstract).
Maier JA; Morelli D; Menard S; Colnaghi MI; Balsaris A
Dipartimento di Scienze e Tecnologie Biomediche-Ospedale San Raffaele, Via Olgettina 58, Milano, Italy
Proc Annu Meet Am Assoc Cancer Res; 37:A123 1996 ISSN 0197-016X

Languages: ENGLISH
Document Type: MEETING ABSTRACTS
Angiogenesis plays a key role in tumor growth, progression and metastasis. To contrast angiogenesis--through induction of endothelial cell death or inhibition of the proliferation--is a potentially useful target for novel forms of anticancer therapy. Endothelial cells undergo apoptosis after withdrawal of growth factors, alteration in the extracellular matrix or exposure to cytokines. We report that tumor necrosis factor alpha (TNF) induces apoptosis of human endothelial cells derived from the umbilical vein (HUVEC) in a dose-dependent fashion. Apoptosis is triggered through a pathway which is independent from the levels of **bcl-2**. On the contrary, TNF stimulates the growth of spontaneously transformed HUVEC (ECV). Indeed, TNF transcriptionally upregulates fibroblast growth factor 1 (FGF-1) expression. The addition of specific FGF-1 **antisense** oligonucleotides inhibits TNF-induced FGF-1 expression and restore the expected responses to TNF: this cytokine inhibits the growth and promotes apoptosis of oligo-**treated** ECV. These results indicate that in ECV endogenous FGF-1 is a potent survival factor capable of antagonizing TNF effects.

4/3,AB/155 (Item 13 from file: 159)
DIALOG(R)File 159:Cancerlit
(c) format only 2001 Dialog Corporation. All rts. reserv.

01147083 97615973
Antisense oligonucleotides in growth factor deprivation therapy enhance expression of **bcl-2** (Meeting abstract).
Rubenstein M; Mirochnik Y; Guinan P
Hektoen Institute, Chicago, IL
Can J Infectious Dis; 6(Suppl C):196C 1995 ISSN 1180-2332
Languages: ENGLISH
Document Type: MEETING ABSTRACTS
The role of **bcl-2** in prostate cancer apoptosis is not well understood, but thought to be involved with the transition toward androgen insensitivity. **Antisense** oligonucleotides (oligos) complementary to mRNA encoding transforming growth factor-alpha (TGF-a) and/or its binding site, the epidermal growth factor receptor (EGFR), inhibit PC-3 tumor cell growth in vitro, and in vivo, providing a model for growth factor deprivation (GFD) therapy of hormone insensitive prostate cancer. To determine whether oligo induced GFD produced similar effects upon **bcl -2** expression as reported following androgen withdrawal, PC-3 cells were **treated** in vitro with oligos against TGF-a and/or EGFR. Cells **treated** with the oligo directed against TGF-a either alone or in combination with that directed against EGFR had increased **bcl-2** expression. This biologic effect observed in surviving cells is similar to that reported following androgen deprivation, providing additional evidence that **antisense** oligos may provide an additional therapy against hormone insensitive tumors based upon GFD.

4/3,AB/156 (Item 14 from file: 159)
DIALOG(R)File 159:Cancerlit
(c) format only 2001 Dialog Corporation. All rts. reserv.

01146108 97609687
Regulatory mechanisms for antigen receptor-mediated B lymphocyte apoptosis (Meeting abstract).
Tsubata T
Department of Medical Chemistry, Faculty of Medicine, Kyoto University
Non-serial; Leukemia and Lymphoma, Pathogenesis and Treatment, Molecular Aspects. 18th Symposium of the International Association for Comparative Research on Leukemia and Related Diseases, p. 172. Kyoto, Japan, October 29-November 3, 1995: 1995
Languages: ENGLISH

Document Type: MEETING ABSTRACTS
Upon interaction with antigens, surface immunoglobulin (sIg) on B cells transduces a signal, which appears to trigger activation and terminal differentiation of mature B cells. However, our and another groups have shown that **treatment** with anti-Ig either in vivo or in vitro induces apoptotic death of mature B cells, suggesting that interaction with antigen results in elimination of B cells. Activation of B cells is thus likely to require, together with an antigen, a costimulatory signal which blocks sIg-mediated apoptosis of mature B cells, as proposed in the two signal model for B cell activation. We have demonstrated that T cell-derived signals through IL-4 receptor and B cell surface antigens CD40 and CD72 block sIg-mediated apoptosis of normal spleen B cells. Moreover, B cell apoptosis by anti-Ig is blocked by T cell-independent antigens such as LPS and dextran sulfate, and mature B cells from autoimmunity-prone New Zealand mice and **bcl-2** transgenic mice are resistant to sIg-mediated apoptosis. Mechanisms for blocking sIg-mediated apoptosis may therefore be required in antibody response to foreign antigens regardless of T-independence or T-dependence and in autoantibody production. When sIg is cross-linked, B lymphoma line WEHI-231 undergoes apoptosis, which is abrogated by CD40 signal. WEHI-231 arrests cell cycle progression prior to apoptosis and the cell cycle arrest is reversed by CD40 signal. When WEHI-231 is **treated** with a suboptimal amount of DNA polymerase inhibitor aphidicolin, CD40 signal no longer blocks sIg-mediated apoptosis. This suggests that cell cycle progression is involved in survival of anti-Ig-**treated** WEHI-231 by CD40 signal. We have demonstrated that cyclin E and a cyclin-dependent kinase inhibitor p27kipl are transcriptionally regulated and are responsible for cell cycle regulation by signals via sIg and CD40 signal. Furthermore, an **antisense** oligonucleotide for p27kipl blocks sIg-mediated apoptosis of WEHI-231. The p27kipl molecule may therefore play an important role in determination whether B cells survive or undergo apoptosis upon interaction with antigens. (4 Refs)

4/3,AB/157 (Item 15 from file: 159)
DIALOG(R)File 159:Cancerlit
(c) format only 2001 Dialog Corporation. All rts. reserv.

01142033 96603099
Antisense oligodeoxynucleotides (ODN) to the **Bcl-2**-JH region downregulate **Bcl-2** expression and inhibit growth of a follicular lymphoma cell line (Meeting abstract).
Abubakr Y; Maki A; Mohammad R; Smith M; Al-Katib A
Div. of Hematology/Oncology, Wayne State Univ. School of Medicine, Detroit, MI
Proc Annu Meet Am Soc Clin Oncol; 14:A1332 1995 ISSN 0732-183X
Languages: ENGLISH
Document Type: MEETING ABSTRACTS
The t(14;18), which is present in 85-90% of the follicular lymphomas, results in overexpression of the **Bcl-2** protein. This protein inhibits apoptosis and plays a key role in lymphomagenesis. The follicular lymphoma cell line WSU-FSCCL has the t(14;18). Previously, we have demonstrated the efficacy of **antisense** ODN directed against the translation initiation site of the **Bcl-2** mRNA against WSU-FSCCL. However, those have the potential to inhibit normal **Bcl-2** expression in hematopoietic stem cells. Unmodified ODN directed to the **Bcl-2**-JH region in the **antisense** (5' CTG AGG AGA CGG TGA CC 3') and sense (5' GGT CAC CGT CTC AG 3') sequences were added to WSU-FSCCL in culture. We found sequence specific inhibition of WSU-FSCCL growth by the **antisense** ODN, that was near maximal at ODN concentration of 40 ug/mL. **Bcl-2** protein expression, as measured by flow cytometry, decreased in the **antisense treated** cells to 30%, as compared to 90% in the control cells. Sense ODN had no significant effects on growth or **Bcl-2** expression. We conclude that **antisense** ODN to the **Bcl-2**-JH region downregulates

Bcl-2 protein expression and inhibits growth of the follicular lymphoma cell line WEHI-FSCCL. These effects were sequence specific. Our results suggest potential utility of these compounds as therapeutic agents for follicular lymphomas. (C) American Society of Clinical Oncology 1997

4/3,AB/158 (Item 16 from file: 159)
DIALOG(R)File 159:Cancerlit
(c) format only 2001 Dialog Corporation. All rts. reserv.

01138592 95609807
Combination of doxorubicin immunoconjugates and molecular intervention in **bcl-2** oncogene expression to overcome drug resistance in small cell lung cancer (Meeting abstract).
Froesch B; Stahel RA; Ludke G; Zangemeister-Wittke U
Div. of Oncology, Dept. of Internal Medicine, Univ. Hosp., Zurich, Switzerland
Proc Annu Meet Am Assoc Cancer Res; 36:A2032 1995 ISSN 0197-016X
Languages: ENGLISH
Document Type: MEETING ABSTRACTS
Drug resistance of small cell lung cancer (SCLC) is the result of a variety of genetic alterations including overexpression of MDR1 and **Bcl-2** . The present study describes two approaches to overcome resistance of SCLC cells to doxorubicin (DOX) **treatment** in vitro. The first was based on the use of monoclonal antibodies (MAbs) to selectively deliver DOX through receptor-mediated endocytosis. For this purpose, DOX was linked to different MAbs that recognize major surface antigens on SCLC cells. Conjugation was accomplished using either a stable linkage or an acid-labile hydrazone bond, which allows release of the drug in the lysosomal compartment. In cell proliferation assays, acid labile DOX-immunoconjugates proved to be highly selective cytotoxic agents against antigen-positive cells with IC50 values ranging between 7 x 10(-7) M and 10(-5) M. They, however, did not overcome the intrinsic drug resistance without the assistance of verapamil. In contrast, resistance was reverted independent of the presence of verapamil using DOX-immunoconjugates carrying a stable linkage. The IC50 values ranged between 7 x 10(-7) M and 10(-6) M, depending on the cell line. The second approach to overcome drug resistance addressed the use of **antisense** oligodeoxynucleotides (ODNs) to counteract expression of **bcl-2**, followed by **treatment** with free or MAb-conjugated DOX. Reduction of **bcl-2** expression with **antisense** ODNs significantly increased the sensitivity of SCLC cells to the **treatment** with DOX delivered in free or in MAb-conjugated form. These data suggest a potential therapeutic use of DOX-immunoconjugates as selective cytotoxic agents in combination with interventions in the expression of genes involved in drug resistance, such as **bcl-2**.

4/3,AB/159 (Item 17 from file: 159)
DIALOG(R)File 159:Cancerlit
(c) format only 2001 Dialog Corporation. All rts. reserv.

01136665 95607880
Estrogen promotes **Bcl-2** proto-oncogene expression in human breast cancer cells (Meeting abstract).
Teixeira C; Pratt MA
Dept. of Pharmacology, University of Ottawa, Ottawa, Ontario, Canada K1H 8M5
Proc Annu Meet Am Assoc Cancer Res; 36:A100 1995 ISSN 0197-016X
Languages: ENGLISH
Document Type: MEETING ABSTRACTS
The **Bcl-2** proto-oncogene suppresses apoptosis induced in a variety of ways. Since estrogen (E2) ablation results in regression of E2-dependent breast tumors we asked whether E2 might alter levels of **Bcl-2** in E2 receptor (ER) positive MCF-7 human breast cancer

cells. MCF-7 cells cultured in the presence of E2 express both the **Bcl-2** mRNA and p26 **Bcl-2**. Depletion of E2 resulted in loss of expression of both mRNA and protein. Reexposure to E2 resulted in profound induction of both **Bcl-2** mRNA and p26 **Bcl-2** within 24 hr. The ER-MDA-MB-231 breast cancer line expressed very low levels of **Bcl-2**. MCF-7 cells **treated** with E2 or expressing the sense orientation of **Bcl-2** in the absence of E2 showed marked resistance to Adriamycin-induced cytotoxicity compared with cells expressing **antisense Bcl-2** or cultured in E2-free medium. We conclude that E2 is able to promote both survival and chemotherapeutic drug resistance in ER+ breast cancer through a mechanism involving the induction of **Bcl-2**.

4/3,AB/160 (Item 18 from file: 159)
DIALOG(R)File 159:Cancerlit
(c) format only 2001 Dialog Corporation. All rts. reserv.

01136631 95607845
Isolation of a drug-sensitizing genetic suppressor element from BCL2 cDNA (Meeting abstract).
Tarasewicz DG; Schott B; Roninson IB
Dept. of Genetics, University of Illinois at Chicago, Chicago, IL 60612-7309
Proc Annu Meet Am Assoc Cancer Res; 36:A65 1995 ISSN 0197-016X
Languages: ENGLISH
Document Type: MEETING ABSTRACTS
The product of the BCL2 oncogene inhibits active cell death (apoptosis) induced by different factors, including anticancer drugs. Since BCL2 is expressed in several types of human cancer, inhibition of this gene represents a potential approach to overcoming tumor resistance to chemotherapy. To develop a specific inhibitor of BCL2, we have used expression selection of genetic suppressor elements, short cDNA segments encoding inhibitory peptides or **antisense** RNAs. A retroviral expression library of DNAasel-generated random fragments of human BCL2 cDNA was introduced into a B-cell leukemia line expressing BCL2 mRNA. Library-transduced cells were **treated** with dexamethasone or vincristine under conditions that induce only low levels of cell death. Cells undergoing apoptosis were isolated by flow sorting, and the transduced BCL2 cDNA fragments were recovered from these cells by PCR. A specific fragment of BCL2 was greatly enriched in apoptotic cells in several independent selections. This fragment encodes a peptide comprising 13% of the BCL2 protein. Both wild-type and mutated versions of this fragment were recovered. When reintroduced into the cells, a mutant fragment encoding the peptide with a single amino acid substitution was found to increase the extent of drug-induced cell death. This effect was abolished by introducing a frameshift mutation near the 5' end of the fragment, indicating that sensitization was mediated by the peptide. Our results suggest the feasibility of developing a peptide inhibitor of BCL2.

4/3,AB/161 (Item 19 from file: 159)
DIALOG(R)File 159:Cancerlit
(c) format only 2001 Dialog Corporation. All rts. reserv.

01061425 96605013
Effectiveness of **bcl-2 antisense** oligodeoxy-nucleotides (AS-ODN) against human follicular small-cleaved cell lymphoma (FSCCL)-SCID mice xenograft model (Meeting abstract).
Abubakr YA; Mohammad R; Maki A; Dan M; Du W; Smith MR; Al-Katib A
Division of Hematology/Oncology and Department of Pathology, Wayne State University, Detroit, MI
Blood; 84(10, Suppl 1):374a 1994 ISSN 0903-1936
Languages: ENGLISH
Document Type: JOURNAL ARTICLE

The t(14:18), seen in 85-90% of follicular lymphomas, is pathogenetically involved in lymphomagenesis through the expression of **bcl-2** and inhibition of apoptosis. We have demonstrated that **bcl-2** AS-ODN downregulates **bcl-2** and inhibits the growth of a human FSCCL line (WSU-FSCCL) in vitro. We have established a human low grade lymphoma xenograft model in SCID mice by injecting WSU-FSCCL cells IV. The animals die after 7 wk due to infiltration of the liver, spleen and bone marrow. Phosphorothioate ODN against the translation initiation site of **bcl-2**-mRNA in the **antisense** (5'-ACC CTG TTC TCC CAG CGT GCG C-3'), and mutated **antisense** (5'-CCC CTT TGC TAC CCG CGG TCG A-3') sequences were administered (10-15 mg/kg/dose/animal) IV or IP to the xenograft model 3 times a week for 2 wk starting on day 7 following tumor injection. Five animals received **antisense**, 4 mutated **antisense** and 5 were untreated controls. **Antisense treated** animals had significantly longer survival, mean 11.6 wk, compared to 7.6 wk for the control, and 7.5 wk for the mutated **antisense treated** animals (P:0.006). More significantly, pathologic examination showed no tumor in the **antisense** group. We conclude that (1) **bcl-2** AS-ODN therapy was effective against FSCCL in the model used; (2) the AS-ODN **treated** animals possibly died of toxicity to normal tissues; and (3) different dose-schedules or AS-ODN to JH-**bcl-2** junction may be tried to prevent/minimize toxicity.

4/3,AB/162 (Item 20 from file: 159)
DIALOG(R)File 159:Cancerlit
(c) format only 2001 Dialog Corporation. All rts. reserv.

01060877 96603606
Regulation of the **Bcl-2** proto-oncogene by estrogen in human breast cancer cells (Meeting abstract).
Pratt C; Teixeira C
Dept. of Pharmacology, Univ. of Ottawa, Ottawa, Ontario, K1H 8M5, Canada
Breast Cancer Res Treat; 32(Suppl):31 1994 ISSN 0167-6806
Languages: ENGLISH
Document Type: JOURNAL ARTICLE
Recent studies have shown that the **Bcl-2** proto-oncogene is able to suppress apoptosis in a large number of cells induced by a variety of conditions. Since estrogen (E2) promotes the growth and survival of human breast cancer cells expressing estrogen receptors (ERs) we asked whether E2 might alter levels of **Bcl-2** in these cells. The ER+ MCF-7 human breast cancer cell line cultured in the presence of E2 expressed both **Bcl-2** mRNA transcript and p26-**Bcl-2** protein. Conversely, the ER- breast cancer line MDA-MB-231 contained extremely low levels of **Bcl-2** mRNA and protein relative to MCF-7 cells. Depletion of estrogen from the medium resulted in loss of expression of the mRNA to almost undetectable levels within 7 days. Reexposure to E2 stimulated a 10-fold induction of transcript levels which were maximal by 24 hours. MCF-7 cells or MCF-7 cells stably expressing a **Bcl-2** **antisense** transcript cultured in E2-free medium were subjected to a 48-hour **treatment** with Adriamycin (10(-7)M). Cytotoxicity assays showed at least a 2-fold increase in cell death compared with Adriamycin **treated** cells supplemented with 10(-8)M E2 or expressing a **Bcl-2** sense transcript. We conclude that E2 is able to promote the survival and contribute to resistance to chemotherapeutic drugs in ER+ breast cancer cells through a mechanism involving the induction of the **Bcl-2** gene.

4/3,AB/163 (Item 21 from file: 159)
DIALOG(R)File 159:Cancerlit
(c) format only 2001 Dialog Corporation. All rts. reserv.

01060380 96600457
Antisense oligodeoxynucleotides to BCR-ABL and apoptosis (Meeting

abstract).
Cotter TG
Dept. of Biology, St. Patricks College, Maynooth, Co. Kildare, Ireland
Non-serial; Gene Therapy: New Frontiers, an International Symposium, September 18-21, 1994, Dublin, Ireland, p.28, 1994: 1994
Languages: ENGLISH
Document Type: JOURNAL ARTICLE
The most common form of cell death under physiological and several pathological conditions is apoptosis (programmed cell death). This cell death process, in contrast to necrosis, is under genetic control and is characterized by cell shrinkage and DNA fragmentation. A number of genes have been identified as playing regulatory roles in apoptosis and these can be divided into two categories: genes that drive the process and those that inhibit. c-Myc and P53 are examples of the former whereas **bcl-2** is an example of a gene that inhibits apoptosis. In the present study we report that the bcr-abl gene acts as an inhibitor of apoptosis in the chronic myelocytic leukaemia cell line K562. **Antisense** oligodeoxynucleotides (10 uM) to the first 15 bases of the bcr portion of the fused gene downregulates expression of the bcr-abl protein. This was demonstrated by both flow cytometry and western blot analysis. Cells depleted of this chimeric protein are considerably more susceptible to the induction of apoptosis by cytotoxic agents such as etoposide, camptothecin and actinomycin D. A nonsense oligodeoxynucleotide control had no effect. **Antisense** by itself did not stimulate apoptosis, but rather sensitized the cells to the induction of apoptosis by cytotoxic agents. Peripheral blood granulocytes from untreated patients with chronic myelocytic leukaemia exhibited a similar inhibition pattern to apoptosis as the K562 cells and this could be over come by **treating** with **antisense**. Thus the combination of **antisense** oligodeoxynucleotides to antiapoptotic genes like bcr-abl together with cytotoxic agents may offer a new therapeutic avenue for the **treatment** of some malignant diseases, where there is deregulated expression of genes that regulate apoptosis.

4/3,AB/164 (Item 22 from file: 159)
DIALOG(R)File 159:Cancerlit
(c) format only 2001 Dialog Corporation. All rts. reserv.

01059234 95611692
Regulation of chemoresistance by **BCL-2** (Meeting abstract).
Reed JC
La Jolla Cancer Research Foundation, La Jolla, CA 92037
Ann Oncol; 5(Suppl 5):A193 1994 ISSN 0923-7534
Languages: ENGLISH
Document Type: JOURNAL ARTICLE
The **bcl-2** gene becomes activated by the t(14;18) chromosomal translocations that occur in the majority of non-Hodgkin's lymphomas, leading to overproduction of the 26-kD **Bcl-2** protein. High levels of **bcl-2** expression can also be found in the absence of detectable structural alterations in the **bcl-2** gene in a wide variety of human cancers including breast, prostate, lung, colorectal and nasopharyngeal carcinomas, neuroblastomas, and some types of leukemia. To explore the molecular basis for the dysregulation of **bcl-2** seen in many types of cancer, we investigated the regulatory elements within the **bcl-2** gene using reporter gene assays. A p53-negative response element was discovered in the **bcl-2** gene, suggesting that p53 loss may result in loss of repression of **bcl-2** gene expression in cancer. Consistent with this hypothesis, expression of a temperature-sensitive version of p53 into a p53-deficient leukemia line resulted in down-regulation of **bcl-2** mRNA and **bcl-2** protein levels upon shift to the permissive temperature, followed by induction of apoptotic cell death. These findings link p53 and **Bcl-2** in a pathway that regulates apoptosis. Previous studies showed that over-production of the **Bcl-2** protein contributes to neoplastic

cell expansion primarily by promoting cell survival through interference with programmed cell death (apoptosis). Because many chemotherapeutic drugs are capable of activating pathways leading to apoptotic cell death, we used gene transfer methods to achieve elevations in the levels of **Bcl-2** protein in various neoplastic lymphoid cell lines as well as a human neuroblastoma line and then tested their relative resistance to killing by several drugs (both cell cycle-dependent and -independent) commonly used in the **treatment** of cancer, including: dexamethasone, methotrexate, Ara-C, VP16, cisplatin, vincristine, 4-HC, 2-CdA, Adriamycin, daunomycin and Taxol. Cells that had been stably infected with a recombinant **bcl-2** retrovirus and that contained 5- to 20-fold elevated levels of **Bcl-2** protein were strikingly more resistant to killing all drugs tested than cells infected with a negative control virus. Conversely, use of **antisense** techniques to reduce levels of **Bcl-2** in t(14;18)-containing lymphoma cell lines resulted in enhanced sensitivity to chemotherapeutic drugs. Though all of these chemotherapeutic drugs were still capable of inducing cell cycle arrest in cells containing high amounts of **Bcl-2** protein, for some drugs surviving cells with high **Bcl-2** were able to re-initiate cell proliferation upon removal of drugs from cultures. Thus, by extending cell survival in the presence of cytotoxic drugs, over-production of the **Bcl-2** protein appears to provide cells with an opportunity to repair drug-induced DNA damage and to resume their proliferative activity, as might occur begin cycles of chemotherapy in clinical scenarios. These findings are consistent with clinical correlative studies that have noted an association between alterations in **bcl-2** gene structure or expression and poor response to therapy in some groups of patients with lymphoma, leukemia, and prostate cancer. Taken together, these findings strongly argue that the relative level of **Bcl-2** protein is an important determinant of the sensitivity of malignant cells to killing by chemotherapeutic drugs, and suggest that methods to reduce **Bcl-2** protein levels or impair **Bcl-2** function could markedly improve the efficacy of conventional antineoplastic drugs in the **treatment** of cancer.

4/3,AB/165 (Item 23 from file: 159)
DIALOG(R)File 159:Cancerlit
(c) format only 2001 Dialog Corporation. All rts. reserv.

00986976 95606905
BCL-2 antisense oligonucleotides suppress t(14;18) B-cell lymphoma growth in a SCID-HU mouse model (Meeting abstract).
Cotter FE; Johnson P; Pocock C; Hall P; Al-Mahdi N; Hawthorn L; Morgan G
LRF Dept. of Hematology and Oncology, Institute of Child Health, London, UK
Non-serial; Fifth International Conference on Malignant Lymphoma, June 9-12, 1993, Lugano, Switzerland, p. 26, 1993.: 1993
Languages: ENGLISH
Document Type: JOURNAL ARTICLE
The t(14;18) translocation is found in 80% of follicular lymphomas and 25% of high-grade B-cell lymphomas. This results in deregulation of the **BCL-2** gene and appears to play a role in oncogenesis. Increments between 5 and 50 x 10(6) cells from a cell line derived from a patient with B-cell lymphoma bearing the t(14;18) translocation were injected by iv, ip and sc routes into a total of 122 SCID mice. The cell line was tested and shown to be negative for the Epstein-Barr virus (EBV). The mice developed lymphoma bearing the t(14;18) translocation with as few as 5 x 10(6) cells within 28 days. This was determined by histological examination. The higher the cell inoculation the more rapidly the lymphoma developed. Engraftment of the tumor cells was determined by PCR for the t(14;18) breakpoint region on peripheral blood samples and could be detected prior to development of overt lymphoma. In addition the t(14;18) translocation within the lymphoma was demonstrated by interphase fluorescence in situ hybridization with chromosome paints to the derivative

14 and 18 translocations derived by chromosome flow sorting. It was possible to passage [redacted] lymphoma from one generation [redacted] the next. Having established a lymphoma model the cells were **treated** in vitro with **antisense** oligonucleotides to the first open-reading frame of the **BCL-2** gene. Control **treatments** with sense and nonsense oligonucleotides were also performed. Cell viability studies were completed and 5 x 10(6) viable cells were inoculated into SCID mice. In vitro sustained downregulation of **BCL-2** was observed after three days of **treatment** with the **antisense** oligonucleotide. At 28 days the sense, nonsense and untreated cell SCID mice had developed lymphoma, however, the **antisense treated** group failed to develop lymphoma suggesting that in vitro downregulation of **BCL-2** leads to an in vivo antilymphoma effect. SCID-hu modelling of B-cell lymphoma bearing the t(14;18) translocation has been demonstrated and the importance of **BCL-2** expression in the lymphoma process suggested. Reduction of the **BCL-2** protein suppresses the oncogenic potential of these lymphoma cells in vivo confirming that it plays an essential role in the development of malignancy.

4/3,AB/166 (Item 24 from file: 159)
DIALOG(R)File 159:Cancerlit
(c) format only 2001 Dialog Corporation. All rts. reserv.

00986939 95606689
Oncogene interactions in apoptosis: Insight in to cancer and the immune system (Meeting abstract).
Green DR
La Jolla Institute for Allergy and Immunology, 11149 N. Torrey Pines Rd. La Jolla, CA 92037
Non-serial; Paris Conference on Apoptosis in AIDS and Cancer, December 2-4, 1993, Paris, France, p. 27 1993.: 1993
Languages: ENGLISH
Document Type: JOURNAL ARTICLE
From a very simplistic point of view, there are four important options available to a cell: it can rest, proliferate, differentiate, or die. If we add the functions of adhesion and locomotion, the coordination and timing of these processes form the basis for development. From a slightly different perspective, they also form the basis of cancer. While an enormous amount of attention has focused on most of these features, the biological importance of cell death, especially apoptosis, has gained general acceptance only in recent years, and significant progress toward the elucidation of the molecular mechanisms that control this form of cell death is now being made. One such mechanism came as something of a surprise: in a number of cases, apoptosis can be promoted by the action of the c-Myc protein, a proto-oncogene product that had previously been associated only with cell proliferation (and presumably, survival). Expression of c-Myc in cells can cause them to enter an apoptotic pathway or proliferate, and this 'decision' depends upon additional signals, such as growth factors that inhibit apoptosis and allow the cells to proliferate. Thus, c-Myc directs cells out of rest and the choice of proliferation or death depends upon the presence or absence of survival signals. Other survival signals are generated by oncogene products that can cooperate with Myc, including **Bcl-2** and Abl. There are implications of this interplay between apoptotic and anti-apoptotic signals in cell transformation and the resistance of some tumor cells to therapeutic agents capable of inducing apoptosis. K562 cells, derived from a patient with CML, express the chimeric Bcr-Abl protein as a result of a chromosomal translocation characteristic of CML. These cells are resistant to the induction of apoptosis by a variety of agents that trigger death in many other cells. To determine whether Bcr-Abl might participate in this anti-apoptotic phenotype, we have employed **antisense** oligodeoxynucleotides to specifically deplete this protein from K562 cells. The **treated** cells now displayed an exquisite susceptibility to apoptosis. Expression of a temperature sensitive v-Abl in these cells

blocked this effect of the **antisense** oligonucleotides (at permissive but not nonpermissive temperature) thus showing that the Abl kinase activity is responsible for the anti-apoptotic effects. Expression of this v-Abl in HL60 similarly produced a resistance to apoptosis induced by a variety of agents, further supporting the participation of this oncogenic protein in establishing an anti-apoptotic pathway in cells. Oncogene interactions therefore that control the life or death of transformed cells also have important roles in normal developmental processes. In the immune system, developing T cells are 'screened' for potential self-reactivity by a process of negative selection, in which activation via the T cell receptor (TCR) induces apoptosis. This process can be mimicked in T cell hybridomas, and appears to depend upon the expression of the c-Myc protein. Evidence that this represents a requirement for the Myc/Max heterodimer will be presented. This evidence is based on the effects of expression of a mutant form of Max that interferes with normal Myc/Max function. As above activated T hybridoma cells appear to be presented with the 'choice' to proliferate or die and the outcome depends not only upon Myc but also other signals. Thus, expression of functional v-Abl kinase in these cells can render them resistant to the activation of apoptosis via TCR ligation. Surprisingly, **Bcl-2** does not appear to be capable of blocking this form of apoptosis, and the reasons for this will provide new insights into the development of the immune system and the process of apoptosis. Although the oncogene products discussed here probably do not directly participate in the biochemical process of apoptosis, they represent molecular controls on this complex mechanism. As such, they give us new ways to look at the life and death of a cell.

4/3,AB/167 (Item 25 from file: 159)
DIALOG(R)File 159:Cancerlit
(c) format only 2001 Dialog Corporation. All rts. reserv.

00986938 95606688
Death genes, anti-death genes and prostate cancer (Meeting abstract).
Buttyan R
Columbia University College of Physicians and Surgeons, NY
Non-serial; Paris Conference on Apoptosis in AIDS and Cancer, December 2-4, 1993, Paris, France, p. 22 1993.: 1993
Languages: ENGLISH
Document Type: JOURNAL ARTICLE
The **treatment** for metastatic prostate cancer generally includes some form of therapy to deplete the male body of testosterone. This **treatment** is initially effective because prostate epithelial cells as well as most prostate cancer cells are dependent on androgens and these cells will undergo apoptosis following castration. If this cell death mechanism was activated in all prostate cells by castration therapy, then it would be curative. However, some fractions of prostate cancer cell inevitably survive and it is this hormone-refractory population of prostate cancer cells that must be targeted if one is to cure metastatic prostate cancer. Since the androgen receptor protein (AR) is needed to transmit hormone signals in the prostate cell, past theories concerning the origin of the hormone-resistant state had always attributed this phenotype to a defect in the expression of the AR protein. Current research fails to provide convincing evidence that lack of or the dysfunction of the AR is the element controlling survival of hormone-resistant prostate cancer cells. In contrast, the research of this laboratory has concentrated on defining the molecular response of prostate cells to androgen withdrawal in order to precisely identify the molecules that are involved in androgen-regulated apoptosis of prostate cells. Experiments have now established that a number of cell-cycle control genes are involved in prostate cell apoptosis. This work has led us to develop a novel and controversial hypothesis in which we describe that the abnormal expression of these cell cycle genes after castration initiates a defective cell cycle in the prostatic epithelial cells. We believe that this 'defective cell cycle' is the suicide process that kills the prostate cell. This research

has also identified at least two potential pathways that can result in the development of hormone resistance in prostate cancer cells without requiring changes in the AR status of the cancer cell. One pathway is mediated by excess expression of the oncoprotein **bcl-2**. The **bcl-2** oncoprotein was the first of the apoptosis-suppressor proteins that was described and we have found that this protein is not expressed in hormone-sensitive prostate epithelium nor in many early invasive (localized) prostate cancer cells. However, every hormone-resistant prostate cancer specimen that we have examined to date expresses high levels of this protein. In vitro studies further establish that **bcl-2** expression can protect prostate cancer cells from a number of apoptotic stimuli. Based on these results, we have proposed that the inappropriate expression of the **bcl-2** oncoprotein is a predominant mechanism by which prostate cancer cells escape hormone dependence. A second pathway may be mediated by mutations in the p53 tumor-suppressor protein. In recent experiments we have found that apoptotic DNA fragmentation can be suppressed by direct therapeutic **treatment** of a regressing rat prostate gland with an **antisense** oligonucleotide that blocks the synthesis of the rat p53 protein. The supposition that p53 expression might be important for hormone-resistance is also supported by the increasing number of laboratories finding that p53 mutations are rare in low-grade prostate cancers but rather common in high-grade and hormone refractory human prostate cancers. Therefore mutations in p53 may be an additional factor regulating prostate cancer's escape from hormone control.

4/3,AB/168 (Item 26 from file: 159)
DIALOG(R)File 159:Cancerlit
(c) format only 2001 Dialog Corporation. All rts. reserv.

00985329 94698631
Antiapoptotic effect of the c-fes proto-oncogene during granulocytic differentiation (Meeting abstract).
Ferrari S; Manfredini R; Grande A; Tagliafico E; Barbieri D; Zucchini P; Citro G; Zupi G; Franceschi C; Torelli U
Inst. of Biological Chemistry, II Medical Clinic, Rome, Italy
Non-serial; International Association for Comparative Research on Leukemia and Related Diseases, 16th Symposium. July 11-16, 1993, Montreal, Quebec, Canada, A25, 1993.: 1993
Languages: ENGLISH
Document Type: JOURNAL ARTICLE
Several oncogenes which encode proteins with tyrosine-kinase activity are involved in proliferation and differentiation of hemopoietic early progenitors. The c-fes proto-oncogene which encodes for a cytoplasmic p92 tyrosine-kinase protein, is expressed at high levels in the terminal stages of granulocytic differentiation, but so far no definite function has been attributed to the product of this oncogene. To tackle this problem, the c-fes proto-oncogene expression has been inhibited in HL60 cells induced to differentiate with retinoic acid (RA) and DMSO. Inhibition of c-fes function was obtained by incubating the cells with a specific **antisense** oligomer complementary to the 5' region of the c-fes mRNA. It was observed that the cells, rather than differentiate, underwent premature cell death showing the morphological and molecular characteristics of apoptosis. Strong support to this conclusion is brought by the observation that GM-CSF and G-CSF promote cell survival in differentiating HL60 cells by interfering with the incubation of c-fes proto-oncogene and thus suppressing apoptosis. The protective effect of G-CSF and GM-CSF is presumably related to events occurring in the short period when the cells, still in cycle, are drown to terminal differentiation. The observation that at 24-48 hr of AS c-fes **treatment** there is a sharp decrease of the proportion of 4c cells suggests that precocious death of cells in S, G2 and M phases of the cell cycle is prevalent in these experimental conditions. The function of the c-fes product may be considered essential to permit the survival of myeloid

cells during differentiation. The function might therefore be similar in granulocytic cells, t[redacted]that of the **bcl-2** oncogene i[redacted]re-B cells. It is possible to conclude that the loss of cell viability that occurs during the in vitro differentiation of myeloid cells, after the complete inhibition of c-fes gene expression and **treatment** with RA-DMSO, is due to activation of programmed cell death rather than an accelerated differentiation.

4/3,AB/169 (Item 27 from file: 159)
DIALOG(R)File 159:Cancerlit
(c) format only 2001 Dialog Corporation. All rts. reserv.

00913210 93689045
Antisense therapeutics.
Bradley MO; Chrisey LA; Hawkins JW
Synthecell Corporation, Rockville, MD 20850
Raven Press Ser Mol Cell Biol; 1:285-93 1992 ISBN 0-88167-854-6
Languages: ENGLISH
Document Type: MONOGRAPH
It is now clear that, in living cells in vitro, appropriately modified **antisense** DNA molecules can decrease the amount or translatability of mRNAs from both cells and infectious organisms. Such decreases in mRNA lead directly to a decrease in the protein coded by that mRNA or to the destruction of an RNA form or critical RNA from a pathogenic organism. Development of **antisense** agents for use against pathogenic and other diseases is discussed, including **antisense** pharmacology, advantages of **antisense** (true rational drug design), disease targets, safety studies (toxicity, pharmacokinetics, genetic toxicity), **antisense** patents, delivery formulation of **antisense** drugs, and manufacture of **antisense** drugs. Only about 15 bases are necessary to bind with absolute precision to any given unique mRNA produced by the human genome. An **antisense** DNA drug should precisely inhibit the synthesis of a targeted protein without disrupting the synthesis of any other protein. Major viral targets for **antisense** agents include HIV, herpes simplex, influenza, and human T-cell leukemia virus. For cancer therapy, **antisense** agents can be designed that can downregulate the expression of oncogenes known to be important in the transformation of a cell line or tumor. For example, expression of the c-fos gene in H-ras-transformed NIH-3T3 cells has been downregulated by using an **antisense** fos construct, causing tumorigenic cells to lose many of their tumorigenic properties. A potential **antisense** approach to cancer therapeutics that would be extremely selective for transformed cells is based on the fact that some malignant cells have specific activating mutations in certain oncogenes such as ras or translocations as in myc or **bcl-2** . These mutations provide a therapeutic window that permits the destruction of cells containing only those specific mutations. Polymerase chain reaction diagnosis of mutations in tumors may provide the necessary genetic information to support this type of therapy. **Antisense** therapeutics are poised to bring a revolution to the pharmaceutical industry. Although unproven, this new technology has the potential to bring to market new drugs with fewer side effects. Novel **treatments** may be devised that utilize gene regulatory circuits to create totally new classes of pharmaceutical agonists that have never been seen before in human therapy. (19 Refs)

4/3,AB/170 (Item 1 from file: 442)
DIALOG(R)File 442:AMA Journals
(c)2001 Amer Med Assn -FARS/DARS apply. All rts. reserv.

00112317
COPYRIGHT American Medical Association 1999

Single-Cell Molecular Biology Implications for the Diagnosis and

Treatment of Neurological Disease (ARTICLE)

O'DELL, DIANNE M.; MCINTOSH, TRACY K.; EBERWINE, JAMES H.
Archives of Neurology
Dec, **1999**; Basic Science Seminars in: tzn1453
LINE COUNT: 00288

The normal functioning of the central nervous system (CNS) requires complex interactions among numerous biological components. The pathophysiology of perturbations in this system is as complex as that of neurological disease. Many methods exist to examine the biological output of dysfunctional cells from a diseased system (eg, immunohistochemical analysis, electrophysiology, and microdialysis), with one goal being to understand the mechanisms of cell death. This understanding may allow the design of therapeutic strategies to prevent cell death and ensuing behavioral abnormalities. Analysis of messenger RNA (mRNA) levels for various genes in CNS tissue may enhance understanding of neurological disease, since cells differ in the complement and abundance of genes they express. One popular method for detecting changes in gene expression is the Northern blot technique, in which total RNA from a sample is extracted and the RNA molecules are separated by size on a denaturing gel and transferred or `blotted' onto nylon membranes that are then probed with radiolabeled DNA for subsequent autoradiograpic detection of gene expression. Arch Neurol. 1999;56:1453-1456

4/3,AB/171 (Item 2 from file: 442)
DIALOG(R)File 442:AMA Journals
(c)2001 Amer Med Assn -FARS/DARS apply. All rts. reserv.

00110299
COPYRIGHT American Medical Association 1999

Targeting in Gene Therapy for Gliomas (ARTICLE)

FUEYO, JUAN; GOMEZ-MANZANO, CANDELARIA; YUNG, W. K. ALFRED; KYRITSIS, ATHANASSIOS P.
Archives of Neurology
Apr, **1999**; Neurological Review: tzn445
LINE COUNT: 00293

Cancer is a disease of a series of genes. Thus, theoretically, brain tumors could be **treated** by targeting their fundamental molecular defects. Currently, most of the approved clinical protocols for gene therapy involve cancer patients. Several of these protocols are designed to improve the **treatment** of brain tumors. In this brief report, we analyze the rationale, advantages, and disadvantages of a series of gene therapy approaches against brain tumors that include transfer of tumor suppressor genes and cell-cycle modulators; suicide or prodrug strategies; immunogene therapy; antiangiogenesis; and oncolytic virus therapy. In summary, in this review, we highlight the translational advances in molecular medicine that broaden our battery of therapies for patients with brain tumors.

4/3,AB/172 (Item 3 from file: 442)
DIALOG(R)File 442:AMA Journals
(c)2001 Amer Med Assn -FARS/DARS apply. All rts. reserv.

00105658
COPYRIGHT American Medical Association 1997

Induction of Heat Shock Protein 70 Protects Thymocytes Against Radiation-Induced Apoptosis (ARTICLE)

GORDON, SHERILYN A.; HOFFMAN, ROSEMARY A.; SIMMONS, RICHARD L.; FORD, HENRI R.
Archives of Surgery
Dec, **1997**; Paper: tzs1277
LINE COUNT: 00478

Objectives: To determine if induction of heat shock protein 70 (HSP 70), a stress protein that plays a cytoprotective role and inhibits cell death in response to various stimuli, will protect thymocytes and T-cell clones from radiation-induced apoptosis, and to define the mechanism of such protection. Design: Thymocytes from BALB/c mice or T-lymphocyte clones were incubated at 43 degreesC for 1 hour to induce HSP 70, then irradiated. Control cells were irradiated but not heated. Fragmentation of DNA was quantitated, and p53, bax, and **bcl-2** expression was analyzed at various times by the Western blot method. Results: Only heated cells expressed HSP 70. The induction of HSP 70 increased basal apoptosis but significantly decreased radiation-induced apoptosis. Furthermore, introduction of an HSP 70 **antisense** oligomer prior to heating reversed the protective effect of HSP 70. Induction of HSP 70 in T-cell clones with sodium arsenite had a similar protective effect against radiation-induced apoptosis. Irradiation induced p53 and markedly up-regulated bax. The expression of p53 peaked at 4 hours and preceded maximal bax induction. Induction of HSP 70 prior to irradiation suppressed p53 and significantly decreased bax levels. Levels of **bcl-2** were unaffected. Conclusions: Our data show that HSP 70 induction protects thymocytes from radiation-induced apoptosis by down-regulating p53 and bax expression. The induction of HSP 70 may represent a novel mechanism by which the immunosuppressive effects and the associated infectious complications of radiation therapy can be minimized. Arch Surg. 1997;132:1277-1282

4/3,AB/173 (Item 4 from file: 442)
DIALOG(R)File 442:AMA Journals
(c)2001 Amer Med Assn -FARS/DARS apply. All rts. reserv.

00103495
COPYRIGHT American Medical Association 1997

Up-regulation of Glial Fibrillary Acidic Protein in the Retina of Primate Eyes With Experimental Glaucoma (ARTICLE)

TANIHARA, HIDENOBU; HANGAI, MASANORI; SAWAGUCHI, SHOICHI; ABE, HARUKI; KAGEYAMA, MASA-AKI; NAKAZAWA, FUMIO; SHIRASAWA, EI-ICHI; HONDA, YOSHIHITO
Archives of Ophthalmology
June, **1997**; Laboratory: tzh752
LINE COUNT: 00394

Objective: To identify molecular mechanisms of retinal responses to intraocular pressure elevation in primate experimental glaucoma. Methods: An experimental glaucoma model was created by repeated laser trabeculophotocoagulation. After the preparation of complementary DNAs from extracted total RNAs in the retinas, polymerase chain reaction (PCR) experiments were performed for the following screening target genes: B-tubulin B2/ and B5/ and glial fibrillary acidic protein (GFAP). To investigate the amplified sequences derived from the PCR experiments, sequencing, subcloning, and Southern blot analysis of PCR products were performed. In addition, an immunohistochemical analysis was performed in an attemptto show the distribution of the target gene products in the retinas. Results: A series of PCR experiments suggested up-regulation of gene expression for GFAP but not for B-tubulins. Sequencing of the PCR products and results of the Southern blot analysis showed that the amplified sequences were derived mainly from the target gene, GFAP, and that increased expression of GFAP was found despite the severity of glaucoma.

Immunohistochemical studies also demonstrated increased expression of GFAP proteins in Muller cells and astrocytes in the retinas of primate eyes with experimental glaucoma. Conclusion: Our study showed up-regulation of GFAP at gene and protein levels, which suggests that glial components in the retina may contribute to the pathologic processes induced by elevated intraocular pressure. Arch Ophthalmol. 1997;115:752-755

4/3,AB/174 (Item 5 from file: 442)
DIALOG(R)File 442:AMA Journals
(c)2001 Amer Med Assn -FARS/DARS apply. All rts. reserv.

00095528
COPYRIGHT American Medical Association 1995

Differential Control of Cell Death in the Skin (ARTICLE)

Archives of Dermatology
Aug, **1995**; Editorial: de_945
LINE COUNT: 00473

4/3,AB/175 (Item 1 from file: 444)
DIALOG(R)File 444:New England Journal of Med.
(c) 2001 Mass. Med. Soc. All rts. reserv.

00114993
Copyright 1995 by the Massachusetts Medical Society

Current Concepts: Chronic Lymphocytic Leukemia (Review Articles)

Rozman, Ciril; Montserrat, Emilio.
The New England Journal of Medicine
Oct 19, **1995**; 333 (16),pp 1052-1057
LINE COUNT: 00382 WORD COUNT: 05282

4/3,AB/176 (Item 2 from file: 444)
DIALOG(R)File 444:New England Journal of Med.
(c) 2001 Mass. Med. Soc. All rts. reserv.

00114617
Copyright 1995 by the Massachusetts Medical Society

Childhood Leukemias (Review Article)

Pui, Ching-Hon.
The New England Journal of Medicine
Jun 15, **1995**; 332 (24),pp 1618-1630
LINE COUNT: 00632 WORD COUNT: 08724

4/3,AB/177 (Item 3 from file: 444)
DIALOG(R)File 444:New England Journal of Med.
(c) 2001 Mass. Med. Soc. All rts. reserv.

00114580
Copyright 1995 by the Massachusetts Medical Society

Mutation of the Androgen-Receptor Gene in Metastatic Androgen-Independent Prostate Cancer (Original Articles)

Taplin, Mary-Ellen; Bubley, Glenn J.; Shuster, Todd D.; Frantz, Martha E.; Spooner, Amy E.; Ogata, George K.; Keer, Harold N.; Balk, Steven P.

The New England Journal of Medicine
May 25, **1995**; 332 (21) pp 1393-1398
LINE COUNT: 00406 WORD COUNT: 05606

Abstract

Background: Metastatic prostate cancer is a leading cause of cancer-related death in men. The rate of response to androgen ablation is high, but most patients relapse as a result of the outgrowth of androgen-independent tumor cells. The androgen receptor, which binds testosterone and stimulates the transcription of androgen-responsive genes, regulates the growth of prostate cells. We analyzed the androgen-receptor genes from samples of metastatic androgen-independent prostate cancers to determine whether mutations in the gene have a role in androgen independence.

Methods: Complementary DNA was synthesized from metastatic prostate cancers in 10 patients with androgen-independent prostate cancer, and the expression of the androgen-receptor gene was estimated by amplification with the polymerase chain reaction. Exons B through H of the gene were cloned, and mutations were identified by DNA sequencing. The functional effects of the mutations were assessed in cells transfected with mutant genes.

Results: All androgen-independent tumors expressed high levels of androgen-receptor gene transcripts, relative to the levels expressed by an androgen-independent prostate-cancer cell line (LNCaP). Point mutations in the androgen-receptor gene were identified in metastatic cells from 5 of the 10 patients examined. One mutation was in the same codon as the mutation found previously in the androgen-independent prostate-cancer cell line. The mutations were not detected in the primary tumors from two of the patients. Functional studies of two of the mutant androgen receptors demonstrated that they could be activated by progesterone and estrogen.

Conclusions: Most metastatic androgen-independent prostate cancers express high levels of androgen-receptor gene transcripts. Mutations in androgen-receptor genes are not uncommon and may provide a selective growth advantage after androgen ablation. (N Engl J Med 1995;332:1393-8.)

4/3,AB/178 (Item 4 from file: 444)
DIALOG(R)File 444:New England Journal of Med.
(c) 2001 Mass. Med. Soc. All rts. reserv.

00112583
Copyright 1994 by the Massachusetts Medical Society

Mechanisms of Disease: Excitatory Amino Acids As A Final Common Pathway For Neurologic Disorders (Review Article)

Lipton, Stuart A.; Rosenberg, Paul A.
The New England Journal of Medicine
Mar 3, **1994**; 330 (9),pp 613-622
LINE COUNT: 00571 WORD COUNT: 07882

4/3,AB/179 (Item 5 from file: 444)
DIALOG(R)File 444:New England Journal of Med.
(c) 2001 Mass. Med. Soc. All rts. reserv.

00112461
Copyright 1994 by the Massachusetts Medical Society

Mechanisms of Disease: The Molecular Basis Of Leukemia (Review Articles)

Cline, Martin J.
The New England Journal of Medicine
Feb 3, **1994**; 330 (5),pp 328-336
LINE COUNT: 00515 WORD COUNT: 07111

4/3,AB/180 (Item 6 from file: 444)
DIALOG(R)File 444:New England Journal of Med.
(c) 2001 Mass. Med. Soc. All rts. reserv.

00109977
Copyright 1992 by the Massachusetts Medical Society

Hodgkin's Disease, Lymphomatoid Papulosis, And Cutaneous T-Cell Lymphoma Derived From A Common T-Cell Clone (Original Articles)

Davis, Thomas H.; Morton, Cynthia C.; Miller-Cassman, Robert; Balk, Steven P.; Kadin, Marshall E.
The New England Journal of Medicine
Apr 23, **1992**; 326 (17),pp 1115-1122
LINE COUNT: 00421 WORD COUNT: 05813

Abstract

Background. Lymphomatoid papulosis is a benign cutaneous eruption that in 10 to 20 percent of patients is associated with the development of lymphoma. The atypical cells of lymphomatoid papulosis histologically resemble the malignant cells of cutaneous T-cell lymphoma or the Reed-Sternberg cells of Hodgkin's disease. We studied a patient in whom lymphomatoid papulosis developed in 1971, Hodgkin's disease in 1975, and cutaneous T-cell lymphoma in 1985, to determine whether these diseases are clonally related.

Methods. The T-cell-receptor alpha-chain gene was cloned and sequenced from a cell line derived from the advanced-stage cutaneous T-cell lymphoma, and the polymerase chain reaction was used to search for this rearrangement of the alpha-chain gene in tissues obtained earlier that were affected by Hodgkin's disease or lymphomatoid papulosis.

Results. The tumor-specific rearrangement of the alpha-chain gene was detected in the patient's earlier tissues affected by lymphomatoid papulosis and Hodgkin's disease, but not in control tissue, including uninvolved tissues from the staging laparotomy for Hodgkin's disease. Cytogenetic studies revealed a translocation, t(8;9)(p22;p24), in cutaneous T-cell lymphoma lines and in a dermatopathic lymph node removed two years before the clinical onset of the cutaneous T-cell lymphoma. Immunohistochemical findings were consistent with an activated T-cell phenotype for the atypical cells of lymphomatoid papulosis, the Reed-Sternberg cells of Hodgkin's disease, and the malignant cells of the T-cell lymphoma.

Conclusions. Lymphomatoid papulosis, Hodgkin's disease, and cutaneous T-cell lymphoma can be derived from a single T-cell clone. A t(8;9) genetic translocation may be involved in the pathogenesis of lymphomatoid papulosis